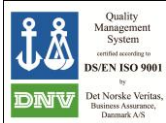





## BIO-SEA

### Performance evaluation in land-based test

Using ultraviolet radiation and a Filtrex filter



This report has been prepared under the DHI Business Management System certified by DNV and specifically for ballast water management system testing certified by Lloyd's Register	
Quality Management	BWMS Testing
ISO 9001	IMO Resolution MEPC.174(58) Annex part 2
	

Approved by
<div style="text-align: right;">13-03-2012</div> <div style="text-align: center;">  </div>
<div style="text-align: center;">  </div>
Approved by
Signed by: Jens Tørsløv

# BIO-SEA

## Performance evaluation in land-based test

Using ultraviolet radiation and a Filtrex filter

Prepared for **BIO-UV**  
Represented by **Ms Charlène Ceresola**



DHI land-based test facility in Hundested

Project No	11809488 (Report No. 1)
Classification	Confidential

Author	Morten Bjergstrøm	

QA unit	Louise Schlüter	

## Contents

<b>Abbreviations.....</b>	<b>1</b>
<b>1 Executive summary .....</b>	<b>2</b>
<b>2 Introduction.....</b>	<b>2</b>
2.1 DHI Ballast Water Centre .....	3
<b>3 Test design .....</b>	<b>4</b>
3.1 Test cycles and trial period.....	4
3.2 Test water and test cycles .....	5
3.2.1 Test water.....	5
3.2.2 Test cycles .....	5
3.2.2.1 First treatment of test water .....	6
3.2.2.2 Second treatment of test water.....	6
3.3 Sampling.....	7
3.3.1 Sample overview .....	7
3.4 Analyses .....	9
3.4.1 Organism size class $\geq 50 \mu\text{m}$ .....	9
3.4.2 Organism size class $\geq 10$ and $< 50 \mu\text{m}$ .....	9
3.4.3 Bacteria.....	9
<b>4 Results.....</b>	<b>9</b>
4.1 Physical/chemical parameters .....	9
4.2 Biological parameters in inlet and control discharge waters.....	10
4.2.1 Live plankton concentrations.....	10
4.2.2 Planktonic diversity in inlet water .....	11
4.2.3 Bacterial concentrations .....	13
4.3 Viable organisms in treated discharge water .....	14
4.3.1.1 Organism size class $\geq 50 \mu\text{m}$ .....	14
4.3.1.2 Organism size class $\geq 10 \mu\text{m}$ and $< 50 \mu\text{m}$ .....	14
4.3.1.3 Bacteria.....	16
4.4 Conclusion on the performance evaluation in the land-based test .....	17
<b>5 Literature.....</b>	<b>18</b>

## Figures

Figure 2.1	DHI Maritime Technology Evaluation Facility .....	3
------------	---	---

## Tables

Table 3.1	Timetable for inlet and discharge operations for all test cycles conducted with the BIO-SEA BWMS. Test cycles B-2 to B-6 were conducted with brackish water, and test cycles M-1 to M-5 were conducted with marine water. The following test cycle pairs were conducted in parallel by use of a common control: B-3 + B-4, M-1 + M-2, M-4 + M-5.....	4
Table 3.2	Target concentrations of DOC, POC and TSS in the test water according to the IMO G8 guidelines. PSU, practical salinity units.....	5
Table 3.3	Target concentrations of live organisms in the test water according to the IMO G8 guidelines.....	5
Table 3.4	Sampling scheme. *Only measurement of primary production .....	7
Table 3.5	Samples from inlet to control and treatment tanks .....	8
Table 3.6	Treated samples collected immediately after first treatment.....	8
Table 3.7	Control and treated discharge samples. Samples for whole effluent toxicity tests were collected during test cycle B-6 (results are reported separately). .....	8

Table 4.1	Brackish water test cycles. Average concentrations of selected parameters: TSS, total suspended solids; DOC, dissolved organic carbon; POC, particulate organic carbon; UV-T, UV-transmittance; PSU, practical salinity units; NR, no requirement.....	9
Table 4.2	Marine water test cycles. Average concentrations of selected parameters: TSS, total suspended solids; DOC, dissolved organic carbon; POC, particulate organic carbon; UV-T, UV-transmittance; PSU, practical salinity units; NR, no requirement. ....	10
Table 4.3	Brackish water test cycles. Live plankton concentrations in the inlet water and in the control discharge water. Organisms in the size class $\geq 10$ and $< 50 \mu\text{m}$ were determined by most probable number (MPN) assay (bold figures) and inverted microscopy (inlet, lower value).....	11
Table 4.4	Marine water test cycles. Live plankton concentrations in the inlet water and in the control discharge water. Organisms in the size class $\geq 10$ and $< 50 \mu\text{m}$ were determined by most probable number (MPN) assay (bold figures) and inverted microscopy (inlet, lower value).....	11
Table 4.5	Brackish water test cycles. Live plankton diversity ( $\geq 50 \mu\text{m}$ ) in the inlet water. ....	12
Table 4.6	Marine water test cycles. Live plankton diversity ( $\geq 50 \mu\text{m}$ ) in the inlet water.....	12
Table 4.7	Brackish water test cycles. Live plankton diversity ( $\geq 10$ and $< 50 \mu\text{m}$ ) in the inlet water.....	13
Table 4.8	Marine water test cycles. Live plankton diversity ( $\geq 10$ and $< 50 \mu\text{m}$ ) in the inlet water. ....	13
Table 4.9	Brackish water test cycles. Bacterial concentrations in the inlet water and the control discharge water. CFU, colony forming units; NR, no requirement. ....	14
Table 4.10	Marine water test cycles. Bacterial concentrations in the inlet water and the control discharge water. CFU, colony forming units; NR, no requirement. ....	14
Table 4.11	Brackish water test cycles. Live plankton concentrations after the first treatment, in treated discharge water after the second treatment and primary production data as additional evidence of treatment efficiency. Organisms in the size class $\geq 10$ and $< 50 \mu\text{m}$ were determined by most probable number (MPN) assay. Data on primary production are expressed as the percentage reduction of the measured primary production in inlet water samples. NR, no requirement. ....	15
Table 4.12	Marine water test cycles. Live plankton concentrations after the first treatment, in treated discharge water after the second treatment and primary production data as additional evidence of treatment efficiency. Organisms in the size class $\geq 10$ and $< 50 \mu\text{m}$ were determined by most probable number (MPN) assay. Data on primary production are expressed as the percentage reduction of the measured primary production in inlet water samples. NR, no requirement. ....	15
Table 4.13	List of algal species capable of growing under the conditions applied in the most probable number (MPN) assay .....	16
Table 4.14	Brackish water test cycles. Bacterial concentrations after the first treatment and in the treated discharge water .....	17
Table 4.15	Marine water test cycles. Bacterial concentrations after the first treatment and in the treated discharge water .....	17

## Appendices

A	QMP and QAPP with Amendments Nos. 1 and 2
B	Data logging from the land-based testing
C	Test results for physical and chemical properties of treated water and biological efficacy analysis in land-based testing
D	Certificate of compliance, ISO 9001 certificate, accreditation and GLP authorisation

## Abbreviations

Abbreviation	Description
BWMS	Ballast water management system
CFU	Colony forming units
DO	Dissolved oxygen
DOC	Dissolved organic carbon
DPM	Disintegrations per minute
FR	Field replicate
IMO	International Maritime Organization
MEPC	Marine Environment Protection Committee
MPN	Most probable number
NR	No requirements
NTU	Nephelometric turbidity units
POC	Particulate organic carbon
PSU	Practical salinity units
QAPP	Quality assurance project plan
QMP	Quality management plan
RQ	Risk quotient
Sd	Standard deviation
T0	Day 0 samples
T5	Day 5 samples
TDI	Tolerable daily intake
TGD	Technical guidance document
TRO	Total residual oxidants
TSS	Total suspended solids
WET	Whole effluent toxicity

## 1 Executive summary

DHI provides independent verification testing services to developers of ballast water management systems at the land-based test facility in Hundested, Denmark. DHI's quality assurance project plan is consistent with the requirements of the International Convention for the Control and Management of Ships Ballast Water and Sediments (IMO 2004).

From September through November 2011, DHI conducted a land-based test of the BIO-UV ballast water management system, BIO-SEA, in accordance with DHI's certification by Lloyd's Register verified by Bureau Veritas. During the series of ten valid test cycles, the ability of the BIO-SEA ballast water management system to (a) successfully treat ballast water without interruption, (b) meet IMO D-2 discharge standards after a five-day retention period and second treatment and (c) discharge water that is environmentally benign (i.e., no residual toxicity) after the five-day retention period and a second treatment, was evaluated.

The BIO-SEA ballast water management system functioned properly during all test cycles and was highly effective at reducing live organism densities in both brackish water and marine water fulfilling the IMO consistent challenge conditions. Live organisms in the size classes defined in the IMO G8 guidelines (MEPC.174(58) (G8)) were discharged at densities below the IMO D-2 standard.

## 2 Introduction

DHI is an independent, international consulting and research organisation with the objectives to advance technological development and competence within the fields of water, environment and health. DHI established a land-based test facility in Hundested, Denmark, in June 2010 with the purpose of providing performance evaluation of ballast water management systems (BWMS) for the approval process. DHI's test facility holds a certificate of compliance issued by Lloyd's Register. The present land-based test was inspected and certified by Bureau Veritas.

DHI has no involvement, intellectual or financial, in the mechanics, design or marketing of the BWMS whose performance has presently been evaluated. To ensure that DHI tests are uncompromised by any real or perceived individual or team bias relative to test outcomes, DHI test activities are subject to rigorous quality assurance, quality control procedures and documentation.

From September through November 2011, DHI conducted a land-based test of the BIO-UV BWMS, BIO-SEA (hereafter BIO-SEA BWMS or BIO-SEA), in accordance with the certification by Lloyd's Register. During the series of ten valid test cycles, the ability of the BIO-SEA BWMS to: (a) successfully treat ballast water without interruption, (b) meet IMO D-2 discharge standards after a five-day retention period and second treatment and (c) discharge water that is environmentally benign (i.e., no residual toxicity) after the five-day retention period and a second treatment, was evaluated.

For an application for final approval, the IMO Convention requires a performance evaluation of the BWMS according to the principles in Resolution MEPC.174(58) (G8) and, when the system is using one or more active substances, Resolution MEPC.169(57) (G9).

The objective of this project was to conduct a land-based test of the BIO-SEA BWMS in accordance with the guidance given in MEPC.174(58) (G8), hereafter referred to as the IMO G8 guidelines.

## 2.1 DHI Ballast Water Centre

The land-based test was carried out by:

DHI  
 Agern Allé 5  
 DK-2970 Hørsholm  
 Denmark

at the land-based test facility:

DHI Maritime Technology Evaluation Facility  
 Færgevejen 18  
 DK-3390 Hundested  
 Denmark

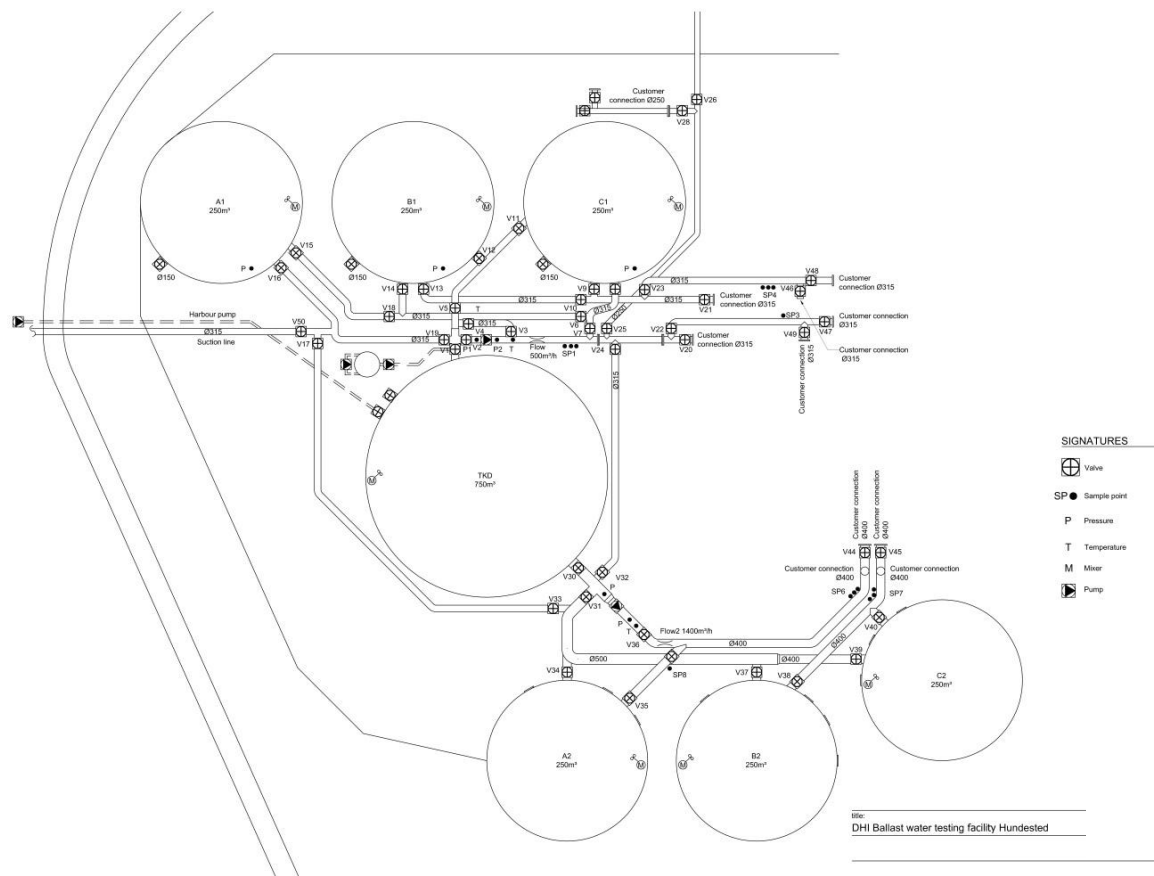


Figure 2.1 DHI Maritime Technology Evaluation Facility

### 3 Test design

#### 3.1 Test cycles and trial period

The BIO-SEA BWMS performance evaluation involved physical and biological characterization of water upon ballasting (inlet of water) and comparison of organisms in control versus treated water immediately following treatment and at discharge after five days storage and second treatment. Biological characterisations supported direct comparison with the IMO D-2 organism categories and standards. During a series of five valid brackish water test cycles (3-32 practical salinity units, PSU) and five valid marine water test cycles (>32 PSU), the BIO-SEA BWMS was tested for its ability to: (a) successfully treat ballast water without interruption, (b) meet IMO D-2 discharge standards after a five-day retention period and second treatment and (c) discharge water that is environmentally benign (i.e., no residual toxicity) after the five-day retention period and a second treatment. A valid test cycle was one, in which the densities of live organisms in the control discharge water were at least 10 times the IMO D-2 standard in accordance with the IMO G8 guidelines.

Table 3.1 Timetable for inlet and discharge operations for all test cycles conducted with the BIO-SEA BWMS. Test cycles B-2 to B-6 were conducted with brackish water, and test cycles M-1 to M-5 were conducted with marine water. The following test cycle pairs were conducted in parallel by use of a common control: B-3 + B-4, M-1 + M-2, M-4 + M-5.

Test cycle identification	Type	Inlet		Discharge		Filter	Number of UV lamps
Brackish water							
B-2	Treatment	2011.09.01	11:47	2011.09.06	10:53	Filtrex	5
	Control		10:35		09:12	-	-
B-3	Treatment	2011.09.08	08:52	2011.09.13	08:46	Filtrex	5
	Control		09:26		09:38	-	-
B-4	Treatment	2011.09.08	11:03	2011.09.13	11:28	Filtrex	4
	Control		09:26		09:38	-	-
B-5	Treatment	2011.10.06	10:19	2011.10.11	10:25	Filtrex	5
	Control		09:16		08:58	-	-
B-6	Treatment	2011.10.13	09:12	2011.10.18	08:09	Filtrex	5
	Control		09:48		08:41	-	-
Marine water							
M-1	Treatment	2011.09.22	09:18	2011.09.27	08:23	Filtrex	5
	Control		09:58		09:02	-	-
M-2	Treatment	2011.09.22	11:19	2011.09.27	10:55	Filtrex	5
	Control		09:58		09:02	-	-
M-3	Treatment	2011.10.20	09:04	2011.10.25	09:49	Filtrex	5
	Control		09:36		08:15	-	-
M-4	Treatment	2011.10.27	09:09	2011.11.01	08:33	Filtrex	5
	Control		09:43		09:13	-	-
M-5	Treatment	2011.10.27	11:16	2011.11.01	10:28	Filtrex	5
	Control		09:43		09:13	-	-

## 3.2 Test water and test cycles

### 3.2.1 Test water

Test water is the term describing the water contained in the source tank of the land-based test facility. According to the IMO G8 guidelines, the land-based tests shall be conducted at a minimum of two different salinities and with the test water qualities described in Tables 3.2 and 3.3.

Table 3.2 Target concentrations of DOC, POC and TSS in the test water according to the IMO G8 guidelines. PSU, practical salinity units

Parameter	Salinity ranges and parameter concentrations		
	>32 PSU	3-32 PSU	<3 PSU
Dissolved organic carbon (DOC)	>1 mg/L	>5 mg/L	>5 mg/L
Particulate organic carbon (POC)	>1 mg/L	>5 mg/L	>5 mg/L
Total suspended solids (TSS)	>1 mg/L	>50 mg/L	>50 mg/L

Table 3.3 Target concentrations of live organisms in the test water according to the IMO G8 guidelines

Parameter	Density	Comment
Organisms $\geq 50 \mu\text{m}$	$>100,000/\text{m}^3$	At least 5 species from at least 3 different phyla/divisions
Organisms $\geq 10 \mu\text{m}$ and $< 50 \mu\text{m}$	$>1,000/\text{mL}$	At least 5 species from at least 3 different phyla/divisions
Heterotrophic bacteria	$>10,000/\text{mL}$	Not further defined

The present land-based test was performed with brackish water (3-32 PSU) and marine water ( $>32$  PSU). The brackish water was collected at the pier outside the test facility in Hundestedt whereas the marine water was prepared by adding salt brine to the brackish water collected at the pier outside the test facility to increase the salinity.

The target concentrations for the test water were accomplished by specific additions if characterisation of the water samples showed that one or more of the parameters specified in Tables 3.2 and 3.3 were deviating from the recommended levels.

If necessary, the concentrations of dissolved organic carbon (DOC), particulate organic carbon (POC) and total suspended solids (TSS = POC + mineral materials) were increased by addition of lignin sulphonate, starch and kaolin, respectively.

If necessary, the numbers of live organisms were increased by additions of harvested indigenous organisms and/or cultures of indigenous organisms or strains of the marine green alga *Tetraselmis suecica* and the marine crustacean *Artemia salina*.

### 3.2.2 Test cycles

A total of ten test cycles, five with brackish water and five with marine water were performed as indicated in Table 3.1. One additional test cycle (test cycle B-1) was performed with 6 UV lamps (see Appendices B and C) The first and second treatment procedures applied in each test cycle are described below.

### 3.2.2.1 First treatment of test water

The first treatment of the test water included the following steps:

- 1 Test water, approx. 200 m<sup>3</sup>, was pumped at a rate of 500 m<sup>3</sup>/h from the source tank through the BWMS for treatment and subsequently into one of the retention tanks
- 2 Test water, approx. 200 m<sup>3</sup>, was pumped at a rate of 300 m<sup>3</sup>/h from the source tank directly into the control tank
- 3 Piping system and sample ports were cleaned.

During ballasting, the flow, pressure, water level, temperature, pH, salinity, turbidity and oxygen level were recorded automatically by use of on-line monitoring.

Samples for documentation of biological performance and water quality before and after treatment were collected.

The treated test water was stored in the retention tank for a minimum of 5 days ± 2 hours after treatment in the BWMS. The same storage time was applied for the test water, which was directed to the control tank without treatment. During storage, both the treated water and control water were stirred slowly.

### 3.2.2.2 Second treatment of test water

- 1 Treated test water (approx. 200 m<sup>3</sup>) contained in the retention tank for 5 days was pumped at a rate of approx. 500 m<sup>3</sup>/h through the BWMS for second treatment, after which it was discharged into the harbour.
- 2 Untreated test water (approx. 200 m<sup>3</sup>) from control tank was discharged into the harbour.
- 3 The retention tanks, piping system and sample ports were cleaned.

During de-ballasting, the flow, pressure, water level, temperature, pH, salinity, turbidity and oxygen concentration were recorded by use of on-line monitoring.

Samples for documentation of biological performance and water quality at discharge were collected.

Furthermore, samples for documentation of control validity and water quality were collected from the control tank at discharge.

### 3.3 Sampling

#### 3.3.1 Sample overview

The type of samples and the points of sampling are summarized in Table 3.4.

Table 3.4 Sampling scheme. \*Only measurement of primary production

Parameter	Ballasting operations			
	Inlet to control and treatment tanks	Inlet to treatment tank after 1 <sup>st</sup> treatment	Discharge from control tank	Discharge from treatment tank after 2 <sup>nd</sup> treatment
Volume	X	X	X	X
Pressure	X	X	X	X
Flow rate	X	X	X	X
Water quality conditions				
Temperature, salinity, turbidity, pH and dissolved oxygen	X	X	X	X
Total suspended solids	X	X	X	X
Dissolved organic carbon and particulate organic carbon	X	-	X	X
Whole effluent toxicity tests	-	-	X	X
Biological diversity and concentrations				
Ambient viable organisms/m <sup>3</sup> ; ≥50 µm	X	X	X	X
Ambient viable organisms/mL; ≥10 and <50 µm	X	X*	X	X
Ambient viable organisms/mL; heterotrophic bacteria; <i>E. coli</i> ; enterococci and <i>Vibrio cholerae</i>	X	X	X	X

The samples and the related analytical parameters are described in Tables 3.5-3.7.

Table 3.5 Samples from inlet to control and treatment tanks

Parameter	SOP No.	Sampled volume (L)
≥10 - <50 µm – primary production	30/1702	10
≥10 - <50 µm – microscopy counting	30/1701	10
≥10 - <50 µm – most probable number (MPN) assay	30/1704	10
≥50 µm – microscopic counting	30/1700	20
<i>E. coli</i> and enterococci	30/1708	0.5
Heterotrophic bacteria	30/1705	0.5
Total suspended solids	30/1768	0.5-2.5
Dissolved organic carbon and particulate organic carbon	30/1769	0.5
Temperature, pH, dissolved oxygen, salinity and turbidity	30/1764	Online

Table 3.6 Treated samples collected immediately after first treatment

Parameter	SOP No.	Sampled volume (L)
≥10 - <50 µm – primary production	30/1702	10
≥50 µm – microscopic counting	30/1700	1,000
<i>E. coli</i> and enterococci	30/1708	0.5
Heterotrophic bacteria	30/1705	0.5
Total suspended solids	30/1768	0.5-2.5
Temperature, pH, dissolved oxygen, salinity and turbidity	30/1764	Online

Table 3.7 Control and treated discharge samples. Samples for whole effluent toxicity tests were collected during test cycle B-6 (results are reported separately).

Parameter	SOP No.	Sampled volume (L)
≥10 - <50 µm – primary production	30/1702	10
≥10 - <50 µm – most probable number (MPN) assay	30/1704	10
≥50 µm – microscopic counting	30/1700	1,000
<i>E. coli</i> and enterococci	30/1708	0.5
<i>Vibrio cholerae</i>	30/1707	0.5
Heterotrophic bacteria	30/1705	0.5
Total suspended solids	30/1768	0.5-2.5
Dissolved organic carbon and particulate organic carbon	30/1769	0.5
Temperature, pH, dissolved oxygen, salinity and turbidity	30/1764	Online
Whole effluent toxicity tests	30/1739	50-100

## 3.4 Analyses

### 3.4.1 Organism size class $\geq 50 \mu\text{m}$

In the samples, the concentrations of viable organisms  $\geq 50 \mu\text{m}$  were determined by using a stereo microscope and a counting chamber. Viable organisms were determined after staining with Neutral Red on the basis of observed mobility. The viable organisms were characterized according to broad taxonomic groups.

### 3.4.2 Organism size class $\geq 10$ and $< 50 \mu\text{m}$

In the inlet water, the concentrations of viable organisms in the size class  $\geq 10$  and  $< 50 \mu\text{m}$  were determined by a combination of two different methods: Analysis of phytoplankton re-growth by use of a most probable number (MPN) assay and inverted microscopy after addition of Lugol's solution. Detailed examination of the algal chloroplasts was applied to confirm that the organisms were alive before the addition of Lugol's solution. The major taxonomic groups present in the sample were noted for each test cycle.

In the control discharge water and the treated discharge water, the concentrations of viable organisms in the size class  $\geq 10$  and  $< 50 \mu\text{m}$  were determined by a combination of two different methods: Analysis of phytoplankton re-growth by use of a most probable number (MPN) assay and measurement of primary production by  $^{14}\text{C}$  incorporation. The MPN assay was consolidated by identification, to the level of taxa or species, of algae that were present in the inlet water and algae that were able to grow under the conditions applied in the MPN assay.

### 3.4.3 Bacteria

Heterotrophic bacteria, *Escherichia coli*, enterococci and *Vibrio cholera*, were analyzed according to internationally accepted methods.

## 4 Results

### 4.1 Physical/chemical parameters

DOC, POC and TSS were in the range of the recommended minimum in the IMO G8 guidelines. The physical/chemical conditions of inlet and discharge waters are summarized in Tables 4.1 and 4.2.

Table 4.1 Brackish water test cycles. Average concentrations of selected parameters: TSS, total suspended solids; DOC, dissolved organic carbon; POC, particulate organic carbon; UV-T, UV-transmittance; PSU, practical salinity units; NR, no requirement.

Parameter	IMO G8 target	Sample	B-2	B-3	B-4	B-5	B-6
TSS (mg/L)	>50	Inlet	55.2	50.3	50.1	57.0	55.4
	NR	Discharge	31.7	26.7	29.1	41.6	42.0
DOC (mg/L)	>5	Inlet	5.54	4.97	4.88	4.73	5.95
	NR	Discharge	5.19	5.02	4.53	4.24	5.37
POC (mg/L)	>5	Inlet	6.23	6.00	6.42	6.13	5.92
	NR	Discharge	2.73	1.96	2.79	3.64	2.76
UV-T (%)	NR	Inlet	59	58	58	54	55
Salinity (PSU)	3-32	Inlet and discharge	17.1	17.2	17.2	17.4	18.0

Table 4.2 Marine water test cycles. Average concentrations of selected parameters: TSS, total suspended solids; DOC, dissolved organic carbon; POC, particulate organic carbon; UV-T, UV-transmittance; PSU, practical salinity units; NR, no requirement.

Parameter	IMO G8 target	Sample	M-1	M-2	M-3	M-4	M-5
TSS (mg/L)	>1	Inlet	12.3	13.5	8.98	9.80	10.2
	NR	Discharge	4.78	5.69	3.86	3.44	3.76
DOC (mg/L)	>1	Inlet	3.23	3.23	3.15	2.28	2.14
	NR	Discharge	3.56	3.57	2.54	2.38	2.37
POC (mg/L)	>1	Inlet	2.82	2.84	3.09	2.77	2.91
	NR	Discharge	0.40	0.47	1.96	0.76	1.02
UV-T (%)	NR	Inlet	80	80	83	87	87
Salinity (PSU)	3-32	Inlet and discharge	35.9	35.9	33.9	33.8	36.4

## 4.2 Biological parameters in inlet and control discharge waters

### 4.2.1 Live plankton concentrations

The densities of live organisms in the inlet water and in the control discharge water were in accordance with the G8 guidelines during all completed test cycles (Tables 4.3 and 4.4).

In the  $\geq 50 \mu\text{m}$  size class, the densities of live organisms in the inlet water ranged from 134,389 to 174,240 organisms/ $\text{m}^3$  in the brackish water test cycles and from 105,569 to 218,333 organisms/ $\text{m}^3$  in the marine water test cycles. The densities of live organisms in the control discharge water ranged from 35,502 to 84,411 organisms/ $\text{m}^3$  in the brackish water test cycles and from 28,319 to 45,129 organisms/ $\text{m}^3$  in the marine water test cycles.

Based on the most probable number (MPN) assay, inlet water concentrations of the smaller planktonic organisms ( $\geq 10$  and  $< 50 \mu\text{m}$ ) ranged from 4,133 to 9,200 organisms/mL in the brackish water test cycles and from 3,133 to 12,467 organisms/mL in the marine water test cycles. The densities of live organisms in the control discharge water were higher than 100 organisms/mL in all test cycles.

Based on inverted microscopy, inlet water concentrations of the smaller planktonic organisms ( $\geq 10$  and  $< 50 \mu\text{m}$ ) ranged from 1,042 to 4,664 organisms/mL in the brackish water test cycles and from 1,794 to 2,189 organisms/mL in the marine water test cycles. The densities of live organisms in the control discharge water were higher than 100 organisms/mL in all test cycles.

Table 4.3 Brackish water test cycles. Live plankton concentrations in the inlet water and in the control discharge water. Organisms in the size class  $\geq 10$  and  $< 50 \mu\text{m}$  were determined by most probable number (MPN) assay (bold figures) and inverted microscopy (inlet, lower value)

Size category	IMO G8 target	Sample	B-2	B-3	B-4	B-5	B-6
$\geq 50 \mu\text{m}$ (organisms/ $\text{m}^3$ )	$> 100,000$	Inlet	134,389	153,529	148,179	174,240	153,011
	$> 100$	Control discharge	51,103	35,502	35,502	73,379	84,411
$\geq 10$ and $< 50 \mu\text{m}$ (organisms/mL)	$> 1,000$	Inlet	<b>8,833</b> 1,116	<b>4,133</b> 4,664	<b>4,133</b> 4,664	<b>7,933</b> 1,914	<b>9,200</b> 1,042
	$> 100$	Control discharge	<b>230</b>	<b>&gt;1,600</b>	<b>&gt;1,600</b>	<b>1,247</b>	<b>&gt;1,600</b>

Table 4.4 Marine water test cycles. Live plankton concentrations in the inlet water and in the control discharge water. Organisms in the size class  $\geq 10$  and  $< 50 \mu\text{m}$  were determined by most probable number (MPN) assay (bold figures) and inverted microscopy (inlet, lower value)

Size category	IMO G8 target	Sample	M-1	M-2	M-3	M-4	M-5
$\geq 50 \mu\text{m}$ (organisms/ $\text{m}^3$ )	$> 100,000$	Inlet	132,311	110,460	105,569	186,922	218,333
	$> 100$	Control discharge	41,459	41,459	28,319	45,129	45,129
$\geq 10$ and $< 50 \mu\text{m}$ (organisms/mL)	$> 1,000$	Inlet	<b>7,567</b> 2,189	<b>7,567</b> 2,189	<b>12,467</b> 2,054	<b>3,133</b> 1,794	<b>3,133</b> 1,794
	$> 100$	Control discharge	<b>603</b>	<b>603</b>	<b>&gt;1,600</b>	<b>&gt;1,600</b>	<b>&gt;1,600</b>

#### 4.2.2 Planktonic diversity in inlet water

The diversity of the plankton in the inlet water was in accordance with the IMO G8 guidelines (5 different species divided between 3 different phyla). Tables 4.5 to 4.8 present an overview of the types of organisms that were identified in the respective test cycles.

Table 4.5 Brackish water test cycles. Live plankton diversity ( $\geq 50 \mu\text{m}$ ) in the inlet water.

Phyla or subphyla	Species	B-2	B-3	B-4	B-5	B-6
Annelida	Polychaete, nereid	X	-	-	-	-
	Spionid polychaete	X	X	X	X	X
Bryozoa	Bryozoa	X	X	X	-	X
Ciliophora	Ciliate	X	-	-	-	-
Crustacea	<i>Acartia clausi</i>	X	X	X	X	X
	<i>Acartia longiremis</i>	-	-	-	-	X
	<i>Artemia</i> sp.	X	X	X	X	X
	<i>Balanus</i> sp.	X	X	X	X	X
	Copepod nauplii	X	X	X	-	-
	Harpacticoid copepod	X	X	X	X	X
	<i>Oithona atlantica</i>	-	-	-	X	X
	<i>Oithona similis</i>	X	X	X	X	X
	<i>Paracalanus parvus</i>	-	-	-	X	X
	<i>Pseudocalanus minitus</i>	-	-	-	X	-
Echinodermata	Pluteus, echinodermata	-	X	X	-	-
Mollusca	Gastropod veliger	X	X	X	-	X
	Mytilus veliger	X	X	X	X	X
Platyhelminthes	Non-segmented worm	X	-	-	-	-
Rotifera	Rotifera, synchaeta	X	X	X	X	X
Sarcomastigophora	<i>Ceratium tripos</i>	X	X	X	X	X

Table 4.6 Marine water test cycles. Live plankton diversity ( $\geq 50 \mu\text{m}$ ) in the inlet water.

Phyla or subphyla	Species	M-1	M-2	M-3	M-4	M-5
Annelida	Spionid polychaete	X	X	X	X	X
Bryozoa	Bryozoa	-	-	X	-	-
Crustacea	<i>Acartia clausi</i>	X	X	X	X	X
	<i>Acartia longiremis</i>	-	-	X	-	-
	<i>Artemia</i> sp.	X	X	X	X	X
	<i>Balanus</i> sp.	X	X	X	X	X
	<i>Centropages hamatus</i>	-	-	X	X	X
	Copepod nauplii	X	X	-	-	-
	Harpacticoid copepod	X	X	-	-	-
	<i>Oithona atlantica</i>	X	X	X	X	X
	<i>Oithona similis</i>	X	X	X	X	X
	<i>Oncaea borealis</i>	-	-	-	X	X
	<i>Paracalanus parvus</i>	-	-	X	X	X
	<i>Pseudocalanus minitus</i>	X	X	-	-	-
Echinodermata	Pluteus, echinodermata	X	X	-	-	-
Mollusca	Mytilus veliger	X	X	-	X	X
Rotifera	<i>Trichocerca marina</i>	-	-	-	X	X
	Rotifera, synchaeta	-	-	X	X	X
Sarcomastigophora	<i>Ceratium tripos</i>	X	X	X	X	X

Table 4.7 Brackish water test cycles. Live plankton diversity ( $\geq 10$  and  $< 50 \mu\text{m}$ ) in the inlet water.

Taxa or groups	Species	B-2	B-3	B-4	B-5	B-6
Bacillariophyceae	<i>Cerataulina pelagica</i>	-	-	-	X	-
	<i>Chaetoceors similis</i>	X	-	-	-	-
	<i>Chaetoceros conmpressus</i>	-	X	X	-	-
	<i>Chaetoceros socialis</i>	X	X	X	X	X
	<i>Guinardia flaccida</i>	X	-	-	-	-
	<i>Leptocylindrus minimus</i>	-	-	-	X	-
	<i>Melosira nummuloides</i>	-	-	-	X	X
	<i>Phaeodactylum tricornutum</i>	-	-	-	X	X
	<i>Rhizosolenia</i> sp.	X	-	-	-	-
	<i>Skeletonema costatum</i>	-	-	-	-	X
Chlorophyceae	<i>Tetraselmis</i> sp.	X	X	X	X	X
Dinophyceae	<i>Ceratium furca</i>	-	-	-	-	X
	<i>Gymnodinium</i> sp.	X	X	X	-	-
	<i>Heterocapsa triquetra</i>	-	-	-	X	X
	<i>Prorocentrum micans</i>	X	-	-	X	X

Table 4.8 Marine water test cycles. Live plankton diversity ( $\geq 10$  and  $< 50 \mu\text{m}$ ) in the inlet water.

Taxa or groups	Species	M-1	M-2	M-3	M-4	M-5
Bacillariophyceae	<i>Cerataulina pelagica</i>	-	-	X	X	X
	<i>Chaetoceros curvisetus</i>	X	X	-	-	-
	<i>Chaetoceros wighamii</i>	-	-	-	X	X
	<i>Leptocylindrus minimus</i>	-	-	X	-	-
	<i>Melosira nummuloides</i>	X	X	X	-	-
	<i>Phaeodactylum tricornutum</i>	-	-	X	X	X
	<i>Pseudonitzschia</i> sp.	-	-	X	-	-
	<i>Rhizosolenia hebetata</i>	-	-	X	-	-
	<i>Skeletonema costatum</i>	-	-	X	X	X
	<i>Thalassiosira nordenskioldii</i>	-	-	-	X	X
	<i>Thalassiosira rotula/gravida</i>	-	-	X	-	-
Chlorophyceae	<i>Tetraselmis</i> sp.	X	X	X	-	-
Cryptophyceae	<i>Cryptophyte</i> sp.	-	-	X	X	X
Dinophyceae	<i>Gymnodinium</i> sp.	-	-	X	-	-
	<i>Heterocapsa triquetra</i>	-	-	X	X	X
	<i>Prorocentrum micans</i>	X	X	-	X	X
	<i>Prorocentrum minima</i>	-	-	-	X	X

#### 4.2.3 Bacterial concentrations

The concentrations of bacteria in the inlet water and the control discharge water are summarized in Tables 4.9 and 4.10.

Table 4.9 Brackish water test cycles. Bacterial concentrations in the inlet water and the control discharge water. CFU, colony forming units; NR, no requirement.

Organism	IMO G8 target	Sample	B-2	B-3	B-4	B-5	B-6
<i>E. coli</i> (CFU/100 mL)	NR	Inlet	7.50	12.17	12.17	35.50	243.33
	NR	Control discharge	16.67	25.33	25.33	80.00	2,420
Enterococci (CFU/100 mL)	NR	Inlet	1,962	492	492	338	328
	NR	Control discharge	959	1,089	1,089	45	58
Heterotrophic bacteria (CFU/mL)	>10,000	Inlet	50,133	33,550	33,550	46,417	41,667
	NR	Control discharge	114,583	236,167	236,167	120,000	>200,000

Table 4.10 Marine water test cycles. Bacterial concentrations in the inlet water and the control discharge water. CFU, colony forming units; NR, no requirement.

Organism	IMO G8 target	Sample	M-1	M-2	M-3	M-4	M-5
<i>E. coli</i> (CFU/100 mL)	NR	Inlet	120.83	120.83	675.33	182.67	182.67
	NR	Control discharge	37.17	37.17	68.83	44.17	44.17
Enterococci (CFU/100 mL)	NR	Inlet	407	407	1,676	722	722
	NR	Control discharge	143	143	2,131	196	196
Heterotrophic bacteria (CFU/mL)	>10,000	Inlet	261,000	261,000	767,500	123,167	123,167
	NR	Control discharge	196,667	196,667	120,417	148,167	148,167

### 4.3 Viable organisms in treated discharge water

The treatment efficiency as defined by the threshold densities of the specific organism categories in IMO D-2 was accomplished during five test cycles with brackish water (3-32 PSU) and five test cycles with marine water (>32 PSU).

#### 4.3.1.1 Organism size class $\geq 50 \mu\text{m}$

The densities of live organism  $\geq 50 \mu\text{m}$  decreased dramatically immediately after first treatment (>99% reduction) in both brackish water and marine water test cycles. The average density of live organisms  $\geq 50 \mu\text{m}$  ranged from 0 to 3.33 individuals per  $\text{m}^3$  in the brackish water test cycles. No live organisms  $\geq 50 \mu\text{m}$  were found in the treated discharge water in any of the marine water test cycles (Tables 4.11 and 4.12).

#### 4.3.1.2 Organism size class $\geq 10 \mu\text{m}$ and $< 50 \mu\text{m}$

In the treated discharge water, the densities of live organisms in the size class  $\geq 10$  and  $< 50 \mu\text{m}$  were consistently below 10 organisms/mL when determined by using the re-growth/MPN assay. The enumeration of algae by use of the MPN assay is directly related to growth, and the MPN values (results) were supported by a substantial reduction of the primary production (Tables 4.11 and 4.12).

Table 4.11 Brackish water test cycles. Live plankton concentrations after the first treatment, in treated discharge water after the second treatment and primary production data as additional evidence of treatment efficiency. Organisms in the size class  $\geq 10$  and  $< 50 \mu\text{m}$  were determined by most probable number (MPN) assay. Data on primary production are expressed as the percentage reduction of the measured primary production in inlet water samples. NR, no requirement.

Size class	IMO G8 target	Sample	B-2	B-3	B-4	B-5	B-6
$\geq 50 \mu\text{m}$ (org./m <sup>3</sup> )	NR	1 <sup>st</sup> treatment	50.5	48.9	30.0	31.7	11.5
	<10	2 <sup>nd</sup> treatment discharge	0.42	3.33	3.33	0.00	2.89
$\geq 10$ and $< 50 \mu\text{m}$ (org./mL)	<10	2 <sup>nd</sup> treatment discharge	<0.18	<0.18	0.19	0.19	0.51
$\geq 10$ and $< 50 \mu\text{m}$ (% red.)	NR	1 <sup>st</sup> treatment Primary production	99.0	98.7	96.9	97.6	97.2
	NR	2 <sup>nd</sup> treatment discharge Primary production	100.0	100.0	100.0	99.7	99.9

Table 4.12 Marine water test cycles. Live plankton concentrations after the first treatment, in treated discharge water after the second treatment and primary production data as additional evidence of treatment efficiency. Organisms in the size class  $\geq 10$  and  $< 50 \mu\text{m}$  were determined by most probable number (MPN) assay. Data on primary production are expressed as the percentage reduction of the measured primary production in inlet water samples. NR, no requirement.

Size class	IMO G8 target	Sample	M-1	M-2	M-3	M-4	M-5
$\geq 50 \mu\text{m}$ (org./m <sup>3</sup> )	NR	1 <sup>st</sup> treatment	17.8	11.1	15.6	15.0	5.83
	<10	2 <sup>nd</sup> treatment discharge	0.00	0.00	0.00	0.00	0.00
$\geq 10$ and $< 50 \mu\text{m}$ (org./mL)	<10	2 <sup>nd</sup> treatment discharge	0.28	0.19	0.19	<0.18	<0.18
$\geq 10$ and $< 50 \mu\text{m}$ (% red.)	NR	1 <sup>st</sup> treatment Primary production	99.2	99.1	99.6	98.3	99.7
	NR	2 <sup>nd</sup> treatment discharge Primary production	100.0	99.9	100.0	99.5	99.7

The ability of algal species to grow is the meaningful definition of viability in an evaluation, of which the target is to determine the efficiency of treatment to reduce the potential of the species in ballast water to proliferate and survive in the natural environment. Both the MPN assay and the measurement of primary production include algae with a dimension of less than  $10 \mu\text{m}$ , which pulls the analyses to the conservative side. On the other hand, measurable growth in the MPN assay depends on the algae's ability to grow under the applied conditions (e.g., nutrients, light and temperature). During the current land-based tests, growth under the conditions applied in the MPN assay has been confirmed for 88% of the algal species found in the brackish inlet water and 82% of the algal species found in the marine inlet water (Table 4.13). These observations show that the conditions in the MPN assay supported growth of more than 82% of the species in the inlet water representing a broad range of algal groups and, therefore, the MPN assay is considered to be a robust measure of viable phytoplankton in the discharge water.

Table 4.13 List of algal species capable of growing under the conditions applied in the most probable number (MPN) assay

Groups	Alga taxa	
Bacillariophyceae	<i>Amphora</i> sp. <i>Attheya decora</i> <i>Cerataulina pelagica</i> <i>Chaetoceros ceratosporus</i> <i>Chaetoceros affinis</i> <i>Chaetoceros compressus</i> <i>Chaetoceros coronatus</i> <i>Chaetoceros debilis</i> <i>Chaetoceros diadema</i> <i>Chaetoceros similis</i> <i>Chaetoceros socialis</i> <i>Chaetoceros wighamii</i> <i>Cylindrotheca closterium/Nitzschia longissima</i> <i>Cymbella</i> spp. <i>Dactyliosira fragilissimus</i> <i>Diploneis</i> sp. <i>Ditylum brightwellii</i> <i>Entomoneis</i> sp.	<i>Fragilaria</i> spp. <i>Gonioceros septentrionalis</i> <i>Grammatophora</i> sp. <i>Guinardia delicatula</i> <i>Leptocylindrus danicus</i> <i>Leptocylindrus minimus</i> <i>Licmophora</i> sp. <i>Melosira nummuloides</i> <i>Navicula</i> spp. <i>Phaeodactylum tricornutum</i> <i>Proboscia alata</i> <i>Pseudonitzschia</i> sp. <i>Skeletonema costatum</i> <i>Synedra</i> sp. <i>Thalassiosira constricta</i> <i>Thalassiosira gravida/rotula</i> <i>Thalassiosira nordenskioldii</i> <i>Thalassiosira</i> spp.
Cryptophyceae	<i>Rhodomonas</i> sp.	cf. <i>Cryptomonas</i> sp.
Dinophyceae	<i>Ceratium furca</i> <i>Dinophysis</i> sp. <i>Gymnodinium</i> sp. cf. <i>Gyrodinium spirale</i> <i>Heterocapsa triquetra</i>	<i>Katodinium glaucum</i> <i>Peridiniella danica</i> <i>Prorocentrum micans</i> <i>Prorocentrum minimum</i>
Euglenophyceae	<i>Eutreptiella</i> sp.	
Chlorophyceae	cf <i>Chlamydomonas</i> cf <i>Pyramimonas</i> sp.	<i>Tetraselmis</i> sp.
Haptophyceae	<i>Haptophyte</i> sp.	cf. <i>Chrysochromulina polylepis</i>
Other	Green flagellates	Macroalgae

#### 4.3.1.3 Bacteria

The densities of live bacteria in the treated discharge water were consistently below the IMO D-2 standard in all test cycles (Tables 4.14 and 4.15).

Table 4.14 Brackish water test cycles. Bacterial concentrations after the first treatment and in the treated discharge water

Organism	IMO G8 target	Sample	B-2	B-3	B-4	B-5	B-6
<i>E. coli</i> (CFU/100 mL)	<250	2 <sup>nd</sup> treatment discharge	<1.00	<1.00	<1.00	<1.00	<1.00
<i>V. cholerae</i> (CFU/100 mL)	<1	2 <sup>nd</sup> treatment discharge	Negative	Negative	Negative	Negative	Negative
Enterococci (CFU/100 mL)	<100	2 <sup>nd</sup> treatment discharge	<1.00	<1.00	<1.00	<1.00	<1.00
Heterotrophic bacteria (CFU/mL)	NR	1 <sup>st</sup> treatment	36.7	162	61.0	20.3	99.8
	NR	2 <sup>nd</sup> treatment discharge	33.7	232	109	32.3	75.0

Table 4.15 Marine water test cycles. Bacterial concentrations after the first treatment and in the treated discharge water

Organism	IMO G8 target	Sample	M-1	M-2	M-3	M-4	M-5
<i>E. coli</i> (CFU/100 mL)	<250	2 <sup>nd</sup> treatment discharge	<1.00	<1.00	<1.00	<1.00	<1.00
<i>V. cholerae</i> (CFU/100 mL)	<1	2 <sup>nd</sup> treatment discharge	Negative	Negative	Negative	Negative	Negative
Enterococci	<100	2 <sup>nd</sup> treatment discharge	<1.00	<1.00	<1.00	<1.00	<1.00
Heterotrophic bacteria (CFU/mL)	NR	1 <sup>st</sup> treatment	109	49.7	361	114	32.7
	NR	2 <sup>nd</sup> treatment discharge	56.7	43.0	26.7	44.0	18.7

#### 4.4 Conclusion on the performance evaluation in the land-based test

The BIO-SEA BWMS functioned properly during the ten test cycles and was highly effective at reducing live organism densities under the applied brackish water and marine water conditions. The densities of live organisms in the size classes and the densities of specific bacteria defined in the IMO G8 guidelines were below the IMO D-2 standard in the treated discharge water in all test cycles with brackish water (3-32 PSU) and in all test cycles with marine water (>32 PSU).

All ten test cycles are considered to fulfil the validity criterion of the IMO G8 guidelines as the organism densities in the control discharges were >100/m<sup>3</sup> (≥50 µm) and >100/mL (≥10 and <50 µm) (MPN assay), respectively.

## 5 Literature

IMO (2004). Adoption of the final act and any instruments, recommendations and resolutions resulting from the work of the conference. International convention for the control and management of ships' ballast water and sediments, BWM/CONF/36 (D2).

IMO (2008). Guidelines for approval of ballast water management systems (G8). Resolution MEPC.174(58).

IMO (2008). Procedure for approval of ballast water management systems that make use of active substances (G9). Resolution MEPC.169(57).



## **A P P E N D I X   A**

QMP and QAPP with Amendments Nos. 1 and 2

# **Quality Management Plan**

## **Performance Evaluation of Ballast Water Management Systems**

**DHI Denmark**

**Version 2.3**

Quality Management Plan  
Performance Evaluation of Ballast Water  
Management Systems  
DHI Denmark  
Version 2.3

Agern Allé 5  
DK-2970 Hørsholm

Tel: +45 4516 9200  
Fax: +45 4516 9292

E-mail: gip@dhigroup.com  
Web: www.dhigroup.com

Project Quality Management Plan Performance Evaluation of Ballast Water Management Systems DHI Denmark Version 2.3		Project No.			
Author Gitte I. Petersen		Date 2011.09.07			
		Approved by Torben Madsen			
2.3	QMP	<i>QIP</i>	<i>TMA</i>	<i>TMA</i>	<i>7/9-11</i>
Revision	Description	By	Checked	Approved	Date
Key words		Classification  <input type="checkbox"/> Open <input type="checkbox"/> Internal <input checked="" type="checkbox"/> Proprietary			

Distribution Client Certification body DHI: /EAT	No. of copies



## CONTENTS

1	TERMS AND ABBREVIATIONS .....	1
2	INTRODUCTION .....	1
3	ORGANISATION .....	2
3.1	Head of department (Torben Madsen) .....	2
3.2	Business area manager (Gitte I. Petersen) .....	3
3.3	Project manager .....	3
3.4	Head of projects.....	4
3.5	Laboratory manager.....	4
4	PERFORMANCE OF PROJECT.....	5
4.1	Agreement .....	5
4.2	Quality Assurance Project Plan (QAPP).....	5
4.3	Services .....	6
4.3.1	Laboratory tests .....	6
4.3.2	Pilot tests .....	6
4.3.3	Land-based tests .....	6
4.3.4	Shipboard tests.....	6
4.4	Reports .....	6
4.4.1	Performance evaluation of BWMS under the IMO convention.....	7
5	QUALITY MANAGEMENT PROCESSES .....	7
5.1	DHI Quality Assurance.....	7
5.2	Document and record control.....	7
5.3	Internal audits .....	8
5.4	Complaint management.....	8
5.5	Subcontractor management.....	8
5.6	Staff competence management .....	8
5.7	Facility management.....	8
5.8	Management review.....	9
6	REFERENCES .....	9

## APPENDICES

A	Standard Operating Procedures (SOPs)
B	Overview of lists
C	Template for Amendments to QAPP
D	Template for Deviations to QAPP



## 1 TERMS AND ABBREVIATIONS

Terms/Abbreviations	Definitions and comments
Active substance	A substance which has a general or specific action on aquatic organisms or bacteria (pathogens)
Ballast Water Management System (BWMS)	A system which removes, renders harmless or avoids uptake or discharge of aquatic organisms and bacteria (pathogens) with ballast water and sediments by mechanical, physical, chemical or biological means acting individually or in combination
Certification Body	Body to certify facilities to conduct performance evaluation of BWMS according to the IMO Convention
Client	The party requesting a performance evaluation of a technology.
Convention	The IMO convention on ballast water
International Maritime Organization (IMO)	United Nations specialized agency with responsibility for the safety and security of shipping and the prevention of marine pollution by ships Comment: IMO has adopted the International Convention for the Control and Management of Ship's Ballast Water and Sediments
Quality Assurance Project Plan (QAPP)	Project-specific technical document describing the BWMS to be tested, the test facility and other conditions affecting the actual design and implementation of the required experiments
Quality Management Plan (QMP)	Generic document describing the quality control management structure and policies of the testing body (including subcontractors and outside laboratories)
Services	The performance evaluation of maritime technologies by laboratory, land-based or shipboard tests or a combination hereof
Standard Operation Procedure (SOP)	Generic document providing rules, guidelines or characteristics for tests or analyses Comment: In-house methods may be used in the absence of a recognized standard, if they are commonly accepted for testing of BWMS or scientifically documented

## 2 INTRODUCTION

The International Maritime Organization (IMO) has adopted the International Convention for the Control and Management of Ship's Ballast Water and Sediments /1/ to reduce the risk of spreading of harmful aquatic organisms and pathogens released with ballast water.

The Convention requires that all ships comply with specified water quality requirements (D2) before ballast water is released into the environment.

The performance evaluation of ballast water management systems (BWMS) aims at documenting compliance with the requirements stated in international guidelines, e.g.:

- Guideline for approval of ballast water management systems - G8 /2/



- Procedure for approval of ballast water management systems that make use of active substances - G9 /3/.

DHI provides services in relation to performance evaluation of maritime technologies and particularly BWMS within the DHI Ballast Water Centre which includes test facilities and laboratories in Denmark and Singapore.

The DHI Ballast Water Centre is organized with a Ballast Water Facility Board including two members from the management in DHI Denmark and two members from the management in DHI Singapore. The object of the Board is to coordinate the development and marketing of services related to the performance evaluation of BWMS within the DHI Group.

The services addressed with the present Quality Management Plan (QMP) include:

- Laboratory tests conducted at the DHI environmental laboratory in Hørsholm, Denmark, aiming at proof-of-concept or technology optimisation
- Pilot-tests conducted at the DHI Maritime Technology Evaluation Facility (hereafter referred to as the “test facility”) in Hundested, Denmark, aiming at technology optimisation
- Land-based tests conducted at the test facility according to international guidelines
- Shipboard tests conducted by DHI Denmark according to international guidelines at vessels, on which the technology is installed

The above activities are collectively referred to as the “services” whereas individual activities are referred to as “projects”.

The aim of the services is to provide independent, third party documentation for the performance of maritime technologies. High quality of the services is ensured through extensive quality management and use of skilled staff.

### **3 ORGANISATION**

#### **3.1 Head of department (Torben Madsen)**

The head of department, business strategy, for DHI’s Department of Environment and Toxicology, has the overall responsibility for the services and the test facility. This includes the following tasks:

- Co-ordination of joint business development between DHI Denmark and DHI Singapore via participation in the Ballast Water Facility Board
- Negotiation of agreements (i.e. service contracts) with clients
- Responsibility for overall co-ordination, planning and costs as required to ensure that the appropriate human resources, facilities and equipment are available for the services
- Appointment of the business area manager, the project manager and task leaders for cross-cutting functions (e.g. production of test water and test facility technical operations)
- Maintenance of the QMP with updated versions as appropriate



- Approval of the Quality Assurance Project Plan (QAPP) and Standard Operation Procedures (SOPs)
- Quality control and approval of test reports (provided that the head of department has not contributed to the technical solution of the project)
- Documentation in relation to
  - Staff training and experience
  - Facilities and their maintenance
  - Records of complaints

### **3.2 Business area manager (*Gitte I. Petersen*)**

The business area manager is responsible for the scientific and technical quality of the services in co-ordination with the head of department. This includes the following managerial tasks:

- Business development and marketing
- Maintenance of generic standards that can serve as formats for drafting the QAPPs and approval of the methods applied in land-based and shipboard tests
- Dialogue with task leaders for cross-cutting functions, e.g. production of test water and test facility technical operations
- Contributions to data interpretation and reporting of land-based and shipboard tests in collaboration with the project manager
- Participation in discussions with the Certification Body on important matters, particularly draft and final reports, together with the project manager
- Co-ordination of the services with the aim to ensure feasibility of parallel projects conducted at the test facility, including decisions related to the functioning of the test facility (e.g. piping and pumps)
- Maintenance of the test facility, connection piping between the test facility and the client's technology, and dialogue with academic and technical staff in order to fulfil DHIs responsibility for operating the test facility during testing
- Quality control of test reports (provided that the business area manager has not contributed to the technical solution of the project)

### **3.3 Project manager**

The project manager is responsible for the management and efficient performance of the project in accordance with the contract between the client and DHI, the QMP and the QAPP.

The project manager's tasks include:

- Organisation and management of the project
- Periodic meetings and other communication with the client to ensure that all necessary information is available in due time
- Preparation of the draft and final QAPP with detailed description of the project, including time schedule and quality assurance of deliverables



- Facilitation of the process for comments and responses to the draft QAPP in dialogue with the client and the Certification Body
- Preparation of amendments and deviations to the QAPP, if any
- Communication of the project time schedule to the Certification Body to enable external audit
- Communication of the QAPP and project time schedule to the internal auditor identified in the QAPP to enable internal audit
- Participation in discussions with the Certification Body on important matters, particularly draft and final reports, together with the business area manager
- Co-ordination and dialogue with the business area manager in relation to safe conditions of work, logistics and technical operations at the test facility
- Co-ordination and dialogue with the laboratory manager in relation to the practical organisation of work involving laboratory technicians; the project manager shall in due time inform the laboratory manager on the types of tests and the required capacity to enable laboratory capacity planning
- Agreements with subcontractors as appropriate for meeting the project deliverables (e.g. chemical analytical laboratory)
- Approval of initiation of the test cycles and interruption of test cycles, e.g. in case of irregularity
- Preparation of reports

### **3.4 Head of projects**

The academic staff (with exception of the business area manager, project manager, task leaders for cross-cutting functions and test co-ordinators) and the secretaries are appointed by the head of projects via dialogue with the business area manager or the project manager as appropriate.

### **3.5 Laboratory manager**

The laboratory manager appoints laboratory technicians for a specific project and allocates tasks to them as part of the laboratory capacity planning. Furthermore, the laboratory manager appoints one or more test co-ordinators among the laboratory technicians or the academic staff for on-site co-ordination of land-based test cycles.

#### **3.5.1.1 Academic staff, laboratory technicians and secretaries**

The tasks of the academic staff, the laboratory technicians and the secretaries include:

- Contributions to the QMP, QAPP and SOPs
- Test co-ordinator function, i.e. co-ordination and keeping timely records of the activities at the test facility during land-based tests
- Sampling at the test facility
- Monitoring of test water quality
- Maintenance of materials and equipment
- Analysis and data processing
- Contributions to test reports



- Archiving of documents and raw data

## **4 PERFORMANCE OF PROJECT**

### **4.1 Agreement**

An agreement between the client and DHI is negotiated and signed according to the DHI manual for project management.

### **4.2 Quality Assurance Project Plan (QAPP)**

The QAPP is a project specific document describing the technology to be tested, the test facility, and other conditions affecting the actual design and implementation of the study. The QAPP is only required for performance evaluation of BWMS in land-based or shipboard tests conducted according to international guidelines.

The QAPP is

- Prepared by the project manager
- Signed by the project manager, the head of department and the internal auditor from the DHI Quality Assurance Unit
- Forwarded to the Certification Body for review and comments
- Forwarded to the client for review, acceptance and signature.

The QAPP typically includes the following titles:

1. Objective
2. Client (including client's monitor, if any)
3. Administration
4. DHI Ballast Water Centre
5. Subcontractors
6. Project management
7. System description
8. Safety handling of active substances
9. Test design (including, for **land-based test**, test cycles, test water, sampling and analyses, and, for **shipboard test**, trial period and locations, sampling and analyses)
10. Validity criteria
11. Pass criteria
12. Time schedule
13. Quality assurance
14. Report
15. Archiving
16. Amendments and deviations, if any
17. References

The QAPP refers to a number of SOPs (see Appendix A).



Amendments and deviations to the QAPP are approved and signed by the project manager. Amendments describe planned changes whereas deviations describe unplanned changes to the QAPP.

The QAPP is subject to internal audit in accordance with the procedures for internal audit of the DHI Quality Management System.

### **4.3 Services**

The project will be conducted as described in the QAPP and subsequent amendments and deviations or, alternatively, as described in the agreement between the client and DHI for projects, for which no QAPP is prepared.

#### **4.3.1 Laboratory tests**

Laboratory tests can be initiated when the BWMS technology is ready for testing and DHI's deliverables are defined. Initiation of testing is decided by the project manager in agreement with the client.

#### **4.3.2 Pilot tests**

Pilot tests can be initiated when the BWMS technology is installed and ready for operation. Initiation of testing is decided in consensus by and between the business area manager and the project manager in agreement with the client.

#### **4.3.3 Land-based tests**

Land-based tests can be initiated when the BWMS technology is installed and ready for operation. Initiation of testing is decided in consensus by and between the business area manager and the project manager in agreement with the client.

The project manager decides when a test cycle in the land-based test is completed and valid, when appropriate by reference to the G8 guidelines /2/, G9 guidelines /3/ or other standards (e.g. US requirements). If required, the project manager can decide to interrupt a test cycle due to technical malfunctioning of the test facility or the BWMS, insufficient state of biological or physical parameters or for other reasons related to the quality of the test water.

#### **4.3.4 Shipboard tests**

Shipboard testing can be initiated when the BWMS technology is installed on the vessel and ready for operation. Initiation of testing is decided by the project manager in agreement with the client.

The project manager decides when a test cycle in the shipboard test is completed and valid by reference to the criteria in G8 /2/ or, if appropriate, to criteria in other standards (e.g. US requirements). If required, the project manager can decide to interrupt a test cycle due to technical malfunctioning of the BWMS, insufficient state of biological or physical parameters or for other reasons related to the water quality.

### **4.4 Reports**

Reports are prepared with the details, format and language described in the agreement between the client and DHI.



#### **4.4.1 Performance evaluation of BWMS under the IMO convention**

For land-based or shipboard tests of BWMS conducted as part of the IMO approval process, the report is typically structured by use of the appropriate headings in the QAPP and shall include a summary of any amendments and deviations to the QAPP.

The report shall include all relevant technical and analytical data and will contain at least the following items:

- Name and address of the client (and monitor, if any)
- Name and address of the testing facility and the dates, on which the test was initiated and completed
- Objectives and procedures stated in the approved QAPP including any changes made to the QAPP
- Results obtained, presented in summarizing tables and as raw data
- Any unforeseen circumstances which may have affected the quality or integrity of the land-based/shipboard testing
- Storage locations of all raw data, the signed QAPP and report
- Descriptions of operations, calculations and transformations performed on the presented data
- Quality assurance statement

The report shall be signed by the project manager, the internal auditor from the DHI Quality Assurance Unit and the head of department.

The final report will be prepared in English and forwarded to the client.

## **5 QUALITY MANAGEMENT PROCESSES**

### **5.1 DHI Quality Assurance**

The services are conducted in accordance with the principles of ISO 9001 by using the DHI Quality Manual and the procedures in this QMP. The Quality Management System of DHI is found compliant with ISO 9001 as part of the ISO 17025 accreditation of the DHI environmental laboratory.

The DHI quality manager is responsible for assigning a trained internal auditor from DHI's Quality Assurance Unit to each project in accordance with the procedures for internal audit of the DHI Quality Management System.

The internal auditor is identified in the QAPP. The internal auditor shall receive the QAPP from the project manager in order to plan and execute internal audit of the project.

### **5.2 Document and record control**

The DHI Quality Manual includes a procedure describing the process of drafting, revising and approving documentation. Standard operation procedures are controlled as described in SOP 30/944.



SOPs 30/921 and 30/937 describe how records of the test are stored, transferred, maintained and controlled in order to ensure data integrity for a period defined in the QAPP, but not shorter than 5 years from completion of the verification.

### **5.3 Internal audits**

Procedure 3 in the DHI Quality System Manual on audit and evaluation and SOP 30/943 describe the process of periodic internal auditing of projects and activities including audit responsibilities and planning, auditor training and competences and audit reporting.

Procedure 4 in DHI Quality System Manual on non-conformities and corrective actions describes how deviations identified during operation and auditing are corrected (corrective actions) and how future occurrence of the same deviations is prevented by improving the quality manual including the process descriptions and working methods (preventive actions).

### **5.4 Complaint management**

Procedure 5 in the DHI Project Management Manual on Complaints describes how complaints are recorded, resolved and reported. If not resolved, complaints are referred to the Certification Body for resolving.

### **5.5 Subcontractor management**

Procedure 4 in the DHI Project Management Manual on subcontractors describes how it is ensured that subcontractors follow quality requirements.

In addition, analytical laboratories providing analyses of any kind should:

- Maintain an ISO 17025 accreditation with the quality management system required herein.
- Apply accredited analytical methods when available.
- Apply other methods according to either international standard methods or in-house methods that are in all cases validated as required for accredited methods.

SOP 30/700 furthermore describes how it is ensured that purchased items such as chemicals and glassware are controlled, accepted and calibrated.

### **5.6 Staff competence management**

Procedure 3 on appraisal interview, post qualifying education and experience in the DHI Employee Conditions Handbook describes how it is ensured that the projects are conducted by staff with adequate competences and knowledge. This is done by maintaining a list of functions in the test process with competence requirements and responsibilities. The list is supported by reference to staff files in the DHI CV database.

### **5.7 Facility management**

SOP 30/945 describes how it is ensured that facilities and equipment are available and fit for the purposes.



## **5.8 Management review**

Procedure 3 of the Quality System Manual on audit and evaluation describes how the DHI management is ensuring that the test centre is working according to this quality manual through mechanisms such as e.g. an annual management review process.

The Quality Manager is responsible for maintenance and development of the quality system and for the internal auditing of all aspects of the system – with daily reference to the Director, Group R&D and Quality Management. The DHI Quality Manual contains rules for reviews of the quality system.

## **6 REFERENCES**

- /1/ IMO (2005): International Convention for the Control and Management of Ships Ballast Water and Sediments. London. International Maritime Organization.
- /2/ MEPC. Guidelines for approval of ballast water management systems (G8). resolution MEPC.174(58). Adopted 10th October 2008.
- /3/ MEPC. Procedure for approval of ballast water management systems that make use of active substances (G9). MEPC.126(53) Adopted 22nd July 2005.



## **A P P E N D I X   A**

### ***BMWS testing-specific Standard Operating Procedures (SOPs)***



SUBJECT/SUBSUBJECT	NO.
ANALYTICAL METHOD ZOOPLANKTON ANALYSIS	30/1700:04
ANALYTICAL METHOD MICROSCOPIC ENUMERATION AND IDENTIFICATION OF MICROALGAE (LUGOL AND CMFDA/FDA)	30/1701:02
ANALYTICAL METHOD DETERMINING PRIMARY PRODUCTION OF MICROALGAE	30/1702:03
ANALYTICAL METHOD DETERMINING DIVERSITY OF MICROALGAL COMMUNITIES BY HPLC ANALYSIS OF PIGMENTS	30/1703:03
ANALYTICAL METHOD DETERMINATION OF VIABLE ALGAE BY MPN	30/1704:02
MICROBIOLOGICAL TESTS DETERMINATION OF TOTAL NUMBER OF BACTERIA BY EPIFLUORESCENCE MICROSCOPY	30/1705:03
MICROBIOLOGICAL TESTS DETERMINATION OF HETEROTROPHIC PLATE COUNT	30/1706:03
MICROBIOLOGICAL TESTS DETERMINATION OF <i>VIBRIO CHOLERA</i> E IN WATER	30/1707:02
MICROBIOLOGICAL TESTS DETERMINATION OF TOTAL COLIFORM, <i>E. COLI</i> AND ENTEROCOCCI Colilert*-18 AND Enterolert-E	30/1708:02
MEASUREMENT METHOD OZONE MEASUREMENT IN WATER	30/1730:02
MEASUREMENT METHOD OZONE MEASUREMENT IN AIR	30/1731:02
MEASUREMENT METHOD TRO MEASUREMENT IN WATER	30/1732:02
HARVESTING, CULTURING AND ADDITION OF ORGANISMS	30/1734:03
COLLECTION OF SEAWATER	30/1735:02
COLLECTION OF FRESH WATER	30/1736:02
CHEMICAL CRITERIA FOR TEST WATER ADDITION OF DOC, POC AND MM	30/1737:02
SAMPLING BIOLOGICAL AND WATER QUALITY PARAMETERS	30/1738:02
SAMPLING WET TEST	30/1739:02
SAMPLING DBP ANALYSIS	30/1740:02
STATISTICS STATISTICAL SURVEILLANCE OF BIOLOGICAL DATA OBTAINED AT TESTS OF BWMSs	30/1760:01
LABELLING SAMPLES COLLECTED AT TEST SITE	30/1761:01
OPERATION OF THE DHI MTEF	30/1762:02
CLEANING RETENTION TANKS, PIPINGS AND OTHER EQUIPMENT AT TEST SITE	30/1763:02
MEASUREMENT METHOD ON-LINE MONITORING OF PRESSURE, TEMPERATURE AND FLOW RATES AT TEST SITE	30/1764:01
MEASUREMENT METHOD FLUORESCENCE	30/1765:02



SUBJECT/SUBSUBJECT	NO.
MEASUREMENT METHOD TURBIDITY	30/1766:03
HEALTH AND SAFETY ENSURING WORKER HEALTH AND SAFETY AT TEST SITE	30/1767:02
MEASUREMENT METHOD DETERMINATION OF TSS	30/1768:02
MEASUREMENT METHOD DETERMINATION OF DOC AND POC	30/1769:02



## ***A P P E N D I X   B***

### ***Overview of lists***



## ***Overview of lists***

The lists mentioned below are kept together with the rest of quality documentation.

### ***Certification Body***

DHI holds a statement describing the Certification Body that has certified the DHI Maritime Technology Evaluation Facility.

### ***List of sub-contractors***

DHI keeps a list of sub-contractors used during the test. The list contains information on name of company, address, contact person, e-mail, telephone number and deliveries.

### ***List of staff approved for functions at the test facility***

DHI keeps a list of persons working at the test facility. The list contains information on the person's activities, responsibility and documentation for training. The person's competence is documented in an available CV.

### ***List of Standard Operation Procedures***

DHI keeps a list of SOPs, including those used in relation to projects conducted at the test facility.



## ***A P P E N D I X   C***

### ***Template for amendments to QAPP***



## AMENDMENT

QAPP DOCUMENT TITLE AND DATE:

AMENDMENT NUMBER:

DATE OF AMENDMENT:

AMENDMENT CONTENTS:

REASON FOR AMENDMENT:

IMPACT OF AMMENDMENT:

PREVENTATIVE ACTION:

If relevant, action to prevent that the same cause of amendment will reoccur in the future.

ORIGINATED BY:

SIGNED BY:

\_\_\_\_\_  
Project manager

\_\_\_\_\_  
DATE

Copy to be sent to the client, the Certification Body and the DHI Quality Assurance Unit.



## ***A P P E N D I X   D***

### ***Template for amendments to QAPP***



## DEVIATION

QAPP DOCUMENT TITLE AND DATE:

DEVIATION NUMBER:

DATE OF DEVIATION:

DESCRIPTION OF DEVIATION:

REASON FOR DEVIATION:

IMPACT OF DEVIATION:

CORRECTIVE ACTION:

If required, actions to be taken to prevent consequences of deviation

ORIGINATED BY:

SIGNED BY:

\_\_\_\_\_  
Project manager

\_\_\_\_\_  
DATE

Copy to be sent to the client, the Certification Body and the DHI Quality Assurance Unit.

## **Quality Assurance Project Plan**

### **Land-based Test of BIO-SEA Ballast Water Management System**



**Quality Assurance Project Plan**  
**Land-based Test of BIO-SEA Ballast Water Management System**

Agern Allé 5  
DK-2970 Hørsholm

Tel: +45 4516 9200  
Fax: +45 4516 9292

E-mail: mbj@dhigroup.com  
Web: www.dhigroup.com

Client BIO UV		Client's representative Charlène Ceresola			
Project Land-based Test of BIO-SEA Ballast Water Management System		Project No. 11809488			
Author Morten Bjergstrøm Gitte I. Petersen		Date 2011.09.01			
		Approved by Torben Madsen			
01	QAPP	mbj	LSC	TMA	1/9.11
Revision	Description	By	Checked	Approved	Date
Key words		Classification			
		<input type="checkbox"/> Open			
		<input type="checkbox"/> Internal			
		<input checked="" type="checkbox"/> Proprietary			
Distribution BIO UV Bureau Veritas DHI: MBJ/GIP/ERA		No. of copies pdf pdf pdf/2			



## CONTENTS

1	OBJECTIVE .....	1
2	CLIENT .....	1
3	CERTIFICATION BODY .....	1
4	DHI BALLAST WATER CENTRE .....	1
4.1	DHI Maritime Technology Evaluation Facility .....	1
4.2	DHI Environmental Laboratory .....	5
5	SUBCONTRACTORS .....	5
6	PROJECT MANAGEMENT .....	5
7	SYSTEM DESCRIPTION .....	5
8	SAFETY HANDLING OF ACTIVE SUBSTANCES .....	5
9	TEST DESIGN .....	5
9.1	Test water .....	5
9.2	Test cycles .....	6
9.2.1	Treatment of test water in the BWMS .....	6
9.2.2	Storage of treated and untreated test water .....	7
9.2.3	Second treatment and discharge of test water .....	7
9.3	Sampling .....	8
9.3.1	Sample overview .....	8
9.3.2	Inlet water and treated water after first treatment .....	9
9.3.3	Treated discharge water and control discharge water .....	9
9.4	Analyses .....	10
9.4.1	Organisms, size class $\geq 50 \mu\text{m}$ .....	10
9.4.2	Organisms, size class $\geq 10 \mu\text{m}$ and $< 50 \mu\text{m}$ .....	10
9.4.3	Bacteria .....	10
9.4.4	On-site measurements .....	11
9.4.5	Chemical analyses .....	11
10	RECORDING OF DATA .....	11
11	VALIDITY CRITERIA .....	11
12	PASS CRITERIA .....	12
13	TIME SCHEDULE .....	12
13.1	Proposed test cycles .....	12
13.2	Reports .....	13



14	QUALITY ASSURANCE .....	13
15	REPORTS .....	13
16	ARCHIVING .....	13
17	AMENDMENTS AND DEVIATIONS .....	14
18	REFERENCES .....	14

## ***APPENDICES***

A	Certificate of Compliance
B	Specification of pumps and other equipment at the test facility
C	Description of the BIO UV ballast water management system BIO-SEA as given by the client
D	Overview of SOPs



## **1 OBJECTIVE**

For an application for final approval, the IMO Convention requires an approval of Ballast Water Management Systems (BWMS) according to the principles laid down in Resolution MEPC.174(58) (G8) /1/ to assure that BWMS approved by administrations are capable of meeting the IMO standard regulation D-2 /2/ in land-based and shipboard evaluations and do not cause unacceptable harm to the vessel, crew, environment or public health.

The objective is to perform a test of the BIO-SEA BWMS (hereafter BIO-SEA) in a land-based test facility according to Resolution MEPC.174(58), Guidelines for approval of ballast water management systems (G8) (hereafter designated the ‘G8 guidelines’).

## **2 CLIENT**

BIO UV  
ZAC de la Petite Carmague  
CS90022  
F-34403 LUNEL Cedex  
France

Contact person: Charlène Ceresola

## **3 CERTIFICATION BODY**

Bureau Veritas  
Division Marine – Direction Technique  
61-67 Boulevard du Château  
F-92200 Neuilly sur Seine  
France

Contact person: Christian Robeson

## **4 DHI BALLAST WATER CENTRE**

### **4.1 DHI Maritime Technology Evaluation Facility**

DHI Maritime Technology Evaluation Facility  
Færgevejen  
DK-3390 Hundested  
Denmark

The DHI Maritime Technology Evaluation Facility (hereafter the ‘test facility’) holds a Certificate of Compliance issued by Lloyd’s Register for the performance of land-based testing of BWMS (Appendix A).



Figure 4.1 DHI Maritime Technology Evaluation Facility, Hundested, Denmark

The test facility is used to conduct biological evaluations of maritime technologies. The test facility is covered by the International Safety Port System (ISPS). Hundested harbour is registered at IMO's website (Port facilities) under Port ID No. 266076DKHUN, Port facility 1651.

The test facility includes seven cylindrical tanks constructed in galvanized steel and coated with a non-toxic top coating:

- One open source tank with a volume of 765 m<sup>3</sup>, Tank D in Figure 4.2 (sometimes described by the approximate volume of 750 m<sup>3</sup>). The source tank is equipped with a propeller which creates a slow circulation in order to maintain the homogeneity of the test water. A bridge across the top of the source tank is established for monitoring the homogeneity.
- Six closed retention tanks, each with a volume of 256 m<sup>3</sup>, Tanks A1, B1, C1, A2, B2 and C2 in Figure 4.2 (sometimes described by the approximate volume of 250 m<sup>3</sup>). Tanks A1 and A2 are also described as 'control tanks' and are used for untreated test water. Tanks B1, C1, B2 and C2 are retention tanks for treated test water.

The piping connecting source tank, control tanks, retention tanks, pump and BWMS is made of polyethylene (diameters 315 mm and 350 mm connecting the A1, B1 and C1 tanks and diameters 400 mm and 500 mm connecting the A2, B2 and C2 tanks).

The piping system connecting the source tank and the retention tanks is equipped with sampling points. The sampling points are equipped with the following sample outlets:

1. Sample outlet for 1-m<sup>3</sup> discharge samples (to be used for analyses of organisms  $\geq 50$   $\mu$ m)
2. Sample outlet for 10-L inlet and 10-L discharge samples (to be used for analyses of organisms  $\geq 10$  -  $< 50$   $\mu$ m) and 20-L inlet samples (to be used for analyses of organisms  $\geq 50$   $\mu$ m)
3. Sample outlet for microbiology samples
4. Sample outlet for samples for analyses of dissolved organic carbon (DOC), particulate organic carbon (POC) and total suspended solids (TSS)



The test facility is equipped with sensors for automatic logging of flow, pressure, water levels, temperature, dissolved oxygen, pH, salinity, conductivity and turbidity.

The test facility includes a main pump with a flow performance of 250-500 m<sup>3</sup>/hour. By use of a harbour piping, this pump can be used to provide a continuous flow of brackish water directly from the harbour to the BWMS with a capacity of up to approx. 300 m<sup>3</sup>/hour. Furthermore, the test facility includes electrical generator power supply up to 150 kVA.

Appropriate volumes of cultivated organisms can, if needed to fulfil the test water quality requirements, be added to the source tank by an auxiliary pump.

The exact configuration of the test facility piping and equipment may be subjects to minor changes.

Specification of pumps and other equipment at the test facility is provided in Appendix B.

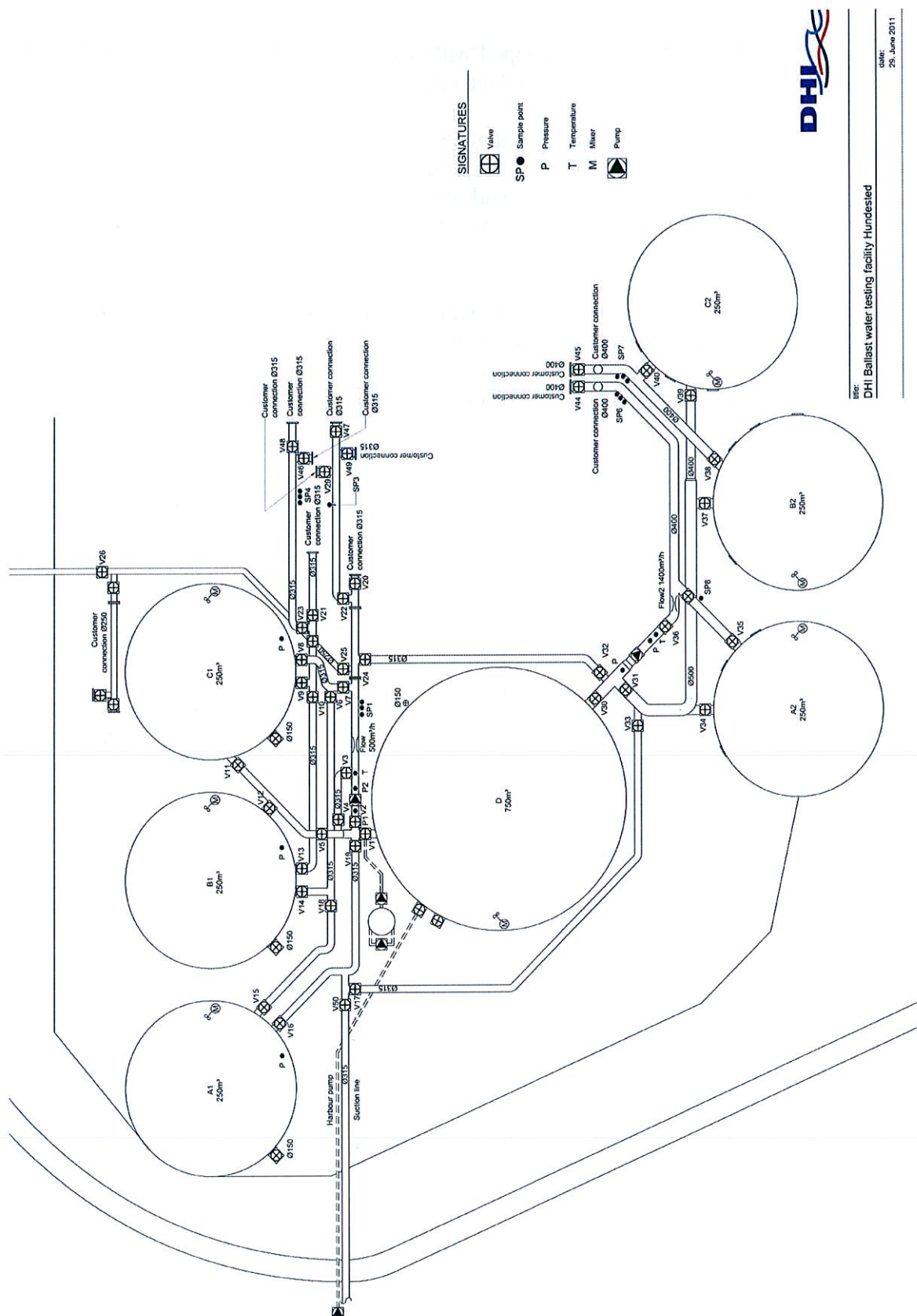


Figure 4.2 DHI Maritime Technology Evaluation Facility, Hundested, Denmark

## **4.2 DHI Environmental Laboratory**

DHI  
Agern Allé 5  
DK-2970 Hørsholm  
Denmark

DHI's environmental laboratory will be used for analyses of samples collected at the test facility. The travel time from the test facility to the laboratory is approx. 50 min, which enables analysis or treatment of the samples within 6 hours.

DHI's environmental laboratory has an accreditation according to ISO 17025 /3/ to perform ecotoxicological tests and is authorized to carry out studies in compliance with the OECD Principles of Good Laboratory Practice (GLP) /4/.

## **5 SUBCONTRACTORS**

The land-based test will be conducted by DHI, and, with the possible exception of verification of *Vibrio cholerae* (according to SOP 30/1707), no subcontractors will be involved.

## **6 PROJECT MANAGEMENT**

The project manager for the study is environmental chemist Morten Bjergstrøm, M.Sc. (environmental chemistry).

## **7 SYSTEM DESCRIPTION**

The description of the BIO-SEA provided by BIO UV is enclosed in Appendix C.

## **8 SAFETY HANDLING OF ACTIVE SUBSTANCES**

No active substances are applied by the BWMS.

## **9 TEST DESIGN**

### **9.1 Test water**

Test water is the term describing the water contained in the source tank. The salinities of the test water (expressed as practical salinity units, PSU) applied in the land-based test will be:

Brackish water (15-25 PSU)  
High saline water (>32 PSU)



When brackish water (15-25 PSU) is used, the source water will be collected immediately south of the pier adjacent to the test facility according to standard operation procedure (SOP) 30/1735; under normal conditions, the natural salinity of the source water will be 15-25 PSU.

When high saline water (>32 PSU) is used, the source water will be collected immediately south of the pier adjacent to the test facility according to SOP 30/1735, and brine will be added to achieve a salinity above 32 PSU.

*Table 9.1 Concentrations of DOC, POC and TSS in the test water according to the G8 guidelines*

Parameter	Salinity		
	> 32 PSU	3-32 PSU	<3 PSU
Dissolved organic carbon (DOC)	> 1 mg/L	> 5 mg/L	> 5 mg/L
Particulate organic carbon (POC)	> 1 mg/L	> 5 mg/L	> 5 mg/L
Total suspended solids (TSS)	> 1 mg/L	> 50 mg/L	> 50 mg/L

*Table 9.2 Concentrations of live organisms in the test water according to the G8 guidelines*

Parameter	Units	Comments
Organisms $\geq 50 \mu\text{m}$	$> 10^5/\text{m}^3$	At least 5 species from at least 3 different phyla/divisions
Organism size: $\geq 10 \mu\text{m} - < 50 \mu\text{m}$	$> 10^3/\text{mL}$	At least 5 species from at least 3 different phyla/divisions
Heterotrophic bacteria	$> 10^4/\text{mL}$	Not further defined

At the time of collection, the water samples will be characterized by use of the appropriate SOPs. If this characterisation shows that the parameters specified in Tables 9.1-9.2 differ from the stated levels, compliance with the requirements will be achieved by specific additions or dilutions.

If necessary, the concentrations of DOC, POC and mineral materials (MM) will be increased by adding, e.g., lignin sulphonate, starch and kaolin clay (SOP 30/1737).

Furthermore, the densities of live organisms will be increased by addition of harvested indigenous organisms and/or cultured species (SOP 30/1734), if necessary.

## 9.2 Test cycles

### 9.2.1 Treatment of test water in the BWMS

The present land-based test will be conducted by use of the source tank (Tank D), control tank (Tank A1) and one retention tank per test cycle (Tank B1 or C1) (Figure 4.2).

The BWMS will be operated by the client during all test cycles. The land-based test will include two (2) sets of test cycles, each consisting of five (5) replicate test cycles with a duration of at least five (5) days. Each set of test cycles will be conducted with test water representing one salinity range.

The following steps will be involved in the treatment of the test water in the BWMS:

1. A fraction of the test water (maximum volume of 250 m<sup>3</sup>) contained in the source tank will be transferred to the BWMS by pumping and treated here, after which it will be transferred to one of the retention tanks.
2. Another fraction of the same test water (maximum volume of 250 m<sup>3</sup>) will be pumped directly into the control tank without passing the BWMS. The untreated test water serves as a control of BWMS performance.
3. Piping system and sample ports will be cleaned (SOP 30/30/1763).

During ballasting, the flow, pressure, temperature, dissolved oxygen, pH, salinity, conductivity, turbidity and water levels in the tanks will be recorded automatically (SOP 30/1764).

Samples will be collected before treatment by use of sampling point 1 (SP1) and after treatment by use of sampling point 2 (SP2). Sampling will be initiated when the flow rate has reached steady-state conditions, i.e. up to 5 minutes from start of operation (SOP 30/1762). Samples will be labelled according to procedures described in SOP 30/1761.

### **9.2.2 Storage of treated and untreated test water**

Following the treatment of the test water in the BWMS, the test water will be stored in the retention tank for at least five days  $\pm$  2 hours. The same storage time will be applied for the test water directed to the control tank without treatment. The retention and the control tanks are equipped with stirrers, and gentle stirring of the treated water and the control water will be applied during storage.

### **9.2.3 Second treatment and discharge of test water**

1. Treated test water contained in the retention tank will be pumped through the BWMS for second treatment, after which it is discharged into the harbour (treated discharge water)
2. Untreated test water contained in the control tank will be discharged into the harbour (control discharge water)
3. The retention tanks, piping system and sample ports will be cleaned (SOP 30/1763)

During de-ballasting, the flow, pressure, water temperature, dissolved oxygen, pH, salinity, conductivity, turbidity and water levels in the tanks will be recorded automatically (SOP 30/1764).

Samples of the treated discharge water will be collected by use of sampling point 2 (SP2) whereas samples of the control discharge water will be collected by use of sampling point 1 (SP1). If appropriate, the samples will be collected by use of approximated isokinetic procedures with fixed sample volumes according to principles described in MEPC.173(58) (G2) /5/ (SOP 30/1738).



## 9.3 Sampling

### 9.3.1 Sample overview

Table 9.3 Overview of sampling and purpose of samples

Parameter	Inlet water to BWMS and control tank (source water)	Treated water, first treatment	Treated discharge water (second treatment)	Control discharge water
<b>Ballasting operations</b>				
Volume	X	X	X	X
Pressure	X	X	X	X
Flow	X	X	X	X
Vendor-specified parameters*	X****	X		X
<b>Water quality conditions</b>				
Temperature, salinity, turbidity, pH, DO**, TSS, DOC***, POC***	X	X	X	X
<b>Concentrations of live organisms</b>				
Ambient viable organisms/m <sup>3</sup> ; ≥50 µm	X	X	X	X
Ambient viable organisms/ml; ≥10 and <50 µm	X	X	X	X
Ambient viable microorganisms/mL; heterotrophic bacteria; <i>E. coli</i> ; enterococci and <i>Vibrio cholerae</i>	X	X	X	X
Whole Effluent Toxicity			X*****	X*****

\* Operational parameters to ensure that the systems have been operated correctly and in accordance with the Operation and Maintenance manual.

\*\* Dissolved oxygen

\*\*\* Measured on inlet and discharge samples

\*\*\*\* Transmittance at 254 nm, 1 cm

\*\*\*\*\* Only at test cycles to be decided later

Triplicate flow-integrated samples will be collected for verification of the G8-D2 requirements. The samples will be stored in thermo boxes with cooler bricks in the dark from the time of collection until handling of the samples in the DHI Environmental Laboratory.



### 9.3.2 Inlet water and treated water after first treatment

Table 9.4 Sampling of inlet water to BWMS and control tank (source water)

Parameter	SOP	Analysis
≥50 µm – Zooplankton microscopy	30/1700	DHI
≥10 - <50 µm - Phytoplankton – MPN assay	30/1704	DHI
≥10 - <50 µm – Phytoplankton microscopy	30/1701	DHI
≥10 - <50 µm – Primary production	30/1702	DHI
<i>E. coli</i> and enterococci	30/1708	DHI
Heterotrophic bacteria	30/1705	DHI
TSS, DOC, POC	30/1768 + 30/1769	DHI
Temperature, pH, O <sub>2</sub> , salinity, turbidity and conductivity	30/1764	DHI
Transmittance at 254 nm, 1 cm		DHI

Table 9.5 Sampling of treated water after first treatment

Parameter	SOP	Analysis
≥ 50 µm – Zooplankton microscopy	30/1700	DHI
≥10 - <50 µm - Primary production	30/1702	DHI
Heterotrophic bacteria	30/1705	DHI
TSS	30/1768 + 30/1769	DHI
Temperature, pH, O <sub>2</sub> , salinity, turbidity and conductivity	30/1764	DHI

### 9.3.3 Treated discharge water and control discharge water

Table 9.6 Sampling of treated discharge water after second treatment

Parameter	SOP	Analysis
≥ 50 µm – Zooplankton microscopy	30/1700	DHI
≥10 - <50 µm - Phytoplankton – MPN assay	30/1704	DHI
≥10 - <50 µm - Primary production	30/1702	DHI
<i>E. coli</i> and enterococci	30/1708	DHI
<i>Vibrio cholerae</i>	30/1707	DHI
TSS, DOC, POC	30/1768 + 30/1769	DHI
Temperature, pH, O <sub>2</sub> , salinity, turbidity and conductivity	30/1764	DHI
Whole Effluent Toxicity*		DHI

\* Only at test cycles to be decided later

Table 9.7 Sampling of control discharge water

Parameter	SOP	Analysis
$\geq 50 \mu\text{m}$ – Zooplankton microscopy	30/1700	DHI
$\geq 10 - < 50 \mu\text{m}$ - Phytoplankton – MPN assay	30/1704	DHI
$\geq 10 - < 50 \mu\text{m}$ - Primary production	30/1702	DHI
<i>E. coli</i> and enterococci	30/1708	DHI
<i>Vibrio cholerae</i>	30/1707	DHI
TSS, DOC, POC	30/1768 + 30/1769	DHI
Temperature, pH, O <sub>2</sub> , salinity, turbidity and conductivity	30/1764	DHI
Whole Effluent Toxicity*		DHI

\* Only at test cycles to be decided later

## 9.4 Analyses

### 9.4.1 Organisms, size class $\geq 50 \mu\text{m}$

In the samples, the concentrations of live zooplankton  $\geq 50 \mu\text{m}$  in minimum dimension will be determined by use of a stereo microscope and a counting chamber according to SOP 30/1700. Viable organisms will be determined on the basis of mobility and morphology and by use of the vital stain Neutral Red. The viable organisms will be characterized according to major taxonomic groups.

### 9.4.2 Organisms, size class $\geq 10 \mu\text{m}$ and $< 50 \mu\text{m}$

In the samples, the concentrations of live phytoplankton in the size class  $\geq 10 - < 50 \mu\text{m}$  in minimum dimension will be analyzed by use of the most probable number (MPN) assay in combination with measurements of primary production. The concentrations of phytoplankton and the presence of taxonomic groups of phytoplanktonic algae in the inlet water will be determined by inverted microscopy. Inverted microscopy will also be used to determine the taxonomic groups of algae that are able to grow under the conditions applied in the MPN assay. The specific methodologies applied are described in SOPs 30/1701, 30/1702 and 30/1704.

The G8 guidelines /1/ define viable organisms as “organisms and any life stages thereof that are living”. There is no commonly accepted criterion to determine the death of algae. The MPN assay with support from the measurements of primary production is considered to provide the most reliable results to be used for a performance evaluation of BWMS because these methods are directly linked to algal growth and, thus, indicative of the ability of the organisms to establish and reproduce in the environment. The MPN assay and the measurement of primary production include algae within the size class  $\geq 10 - < 50 \mu\text{m}$  and algae that are less than  $< 10 \mu\text{m}$ .

### 9.4.3 Bacteria

The concentrations of heterotrophic bacteria will be determined according to SOP 30/1706 (ISO 6222). *E. coli* and enterococci will be analyzed according to SOP 30/1708. *Vibrio cholerae* will be analyzed according to the method described in SOP 30/1707.



#### **9.4.4 On-site measurements**

The measurements conducted at the test facility according to SOPs 30/1764 and 30/1766 will include:

- pH
- Temperature
- Salinity
- Conductivity
- Turbidity
- Dissolved oxygen
- Ballast system pressure
- Ballast system flow rates

#### **9.4.5 Chemical analyses**

Chemical analyses conducted at the DHI Environmental Laboratory will include the following measurements:

- TSS
- POC
- DOC
- Transmittance at 254 nm, 1 cm

## **10 RECORDING OF DATA**

During the test, logging of data related to

- Personnel working at the test facility
- Operational procedures and monitoring
- Sampling and analyses

is performed in accordance with the data logging procedures described in the respective SOPs.

An overview of the respective SOPs used during performance of the land-based test is presented in Appendix D.

## **11 VALIDITY CRITERIA**

A test cycle will be regarded as valid if the average results of the untreated test water (control test water) at the time of discharge are equal to or higher than 10 times the values stated in Regulation D-2.1 /2/, i.e.:

- $\geq 100$  organisms/m<sup>3</sup> for the organisms size class  $\geq 50$   $\mu$ m
- $\geq 100$  organisms/mL for the organisms size class  $\geq 10$   $\mu$ m to  $< 50$   $\mu$ m.



## 12 PASS CRITERIA

The pass criteria for the land-based test cycles are:

1. The test cycle shall be valid according to the validity criteria
2. The average density of organisms larger than or equal to 50  $\mu\text{m}$  in minimum diameter in the replicate samples shall be less than 10 viable organisms per  $\text{m}^3$  at discharge
3. The average density of organisms smaller than 50  $\mu\text{m}$  and larger than or equal to 10  $\mu\text{m}$  in minimum diameter in the replicate samples shall be less than 10 viable organisms per mL at discharge, based on the combined results from the MPN analysis, viability counts and levels of primary production
4. The average density of *Vibrio cholerae* (serotypes O1 and O139) shall be less than 1 colony forming unit (CFU) per 100 mL at discharge
5. The average density of *E. coli* in the replicate samples shall be less than 250 CFU per 100 mL at discharge
6. The average density of intestinal enterococci in the replicate samples shall be less than 100 CFU per 100 mL at discharge

## 13 TIME SCHEDULE

### 13.1 Proposed test cycles

Table 13.1 Proposed time schedule for land-based test cycles

Month	September			October		November		
Week No.	35	36	38	40	42	44	45	46
Test cycle No.	X (1) X+1	2 3	4 5	Sup1	6 7	8 9	10	Sup2

Green: Brackish water test cycles  
 Blue: High saline water test cycles  
 X: Different UV configurations  
 Sup: Supplementary test

In week 35, two test cycles with different UV configurations will be conducted. Two supplementary test cycles with a second filter will be performed.

DHI decides the applied test water salinities and the timing of the test of the test cycles within the access period.

Dependent on weather conditions and the possibilities of preparing the right test water quality, the land-based test cycles will be conducted as scheduled above.

Ballasting is performed on Thursdays from approx. 08:00 am and the de-ballasting is performed on the following Tuesday from approx. 08:00 am, if at all practicable.



### **13.2 Reports**

Separate reports will be prepared for BIO-SEA with FILTREX filter and for BIO-SEA with Filtersafe filter:

- Draft final reports will be prepared within ten (10) weeks after the successful completion of the ten or two test cycles, respectively
- Final reports will be issued three weeks after the client's approval of the relevant draft report

## **14 QUALITY ASSURANCE**

The policy, scope, organisation, responsibilities, principles for performing the test and quality management process are described in the Quality Management Plan /6/. The Quality Management Plan has been prepared in accordance with the principles of ISO 9001 and in compliance with ISO 17025.

The DHI Quality Assurance Unit will review the Quality Assurance Project Plan and conduct inspections of the test cycles, the analyses and the raw data.

The final report will be audited.

Inspection and audit will be carried out by Quality Assurance personnel independent of the staff involved in the land-based testing.

## **15 REPORTS**

Separate reports will be prepared for BIO-SEA with FILTREX filter and for BIO-SEA with Filtersafe filter. The following reports will be prepared:

- Draft final reports compiling all relevant data from the ten/two test cycles, data interpretation and conclusion
- Final reports

## **16 ARCHIVING**

All data generated and all other records and information relevant to the quality and integrity of the land-based testing will be retained in the archives of DHI for a period of five years after issue of the final report.

## **17 AMENDMENTS AND DEVIATIONS**

Amendments are planned changes to the Quality Assurance Project Plan. Deviations are unplanned changes. Amendments and deviations will be signed by the project manager and documented in the file and the final report.

## **18 REFERENCES**

- /1/ Resolution MEPC.174(58). Adopted on 10 October 2008. Guidelines for approval of ballast water management systems (G8)
- /2/ IMO (2004): Adoption of the final act and any instruments, recommendations and resolutions resulting from the work of the conference. International convention for the control and management of ships' ballast water and sediments (BWM/CONF/36).
- /3/ EN ISO/IEC 17025 (2005): General requirements for the competence of testing and calibration laboratories /ISO/IEC 17025:2005).
- /4/ OECD Principles of Good Laboratory Practice (as revised in 1997). Organisation for Economic Co-operation and Development (OECD), Paris. ENV/MC/CHEM (98)17.
- /5/ Resolution MEPC.173(58). Adopted on 10 October 2008. Guidelines for approval of ballast water sampling (G2)
- /6/ Quality Management Plan (QMP) for DHI Maritime Technology Evaluation Facility (MTEF). March 2011.



## APPROVAL OF QUALITY ASSURANCE PROJECT PLAN

DHI

Project management

  
Morten Bjergstrøm

Date: 1/9-11

DHI management

  
Torben Madsen

Date: 1/9-11


Quality Assurance Unit

  
Louise Schlüter

Date: 1/9-11

This QAPP is accepted and my signature authorizes the study to proceed as described in this document.

Client

  
Charlène Ceresola  
BIO UV

Date: 02/09/2011



## ***A P P E N D I X   A***

### ***Certificate of Compliance***



Certificate no: **DS/I093222**  
Page 1 of 1



## Certificate of Compliance

Office: **Lloyd's Register EMEA**  
Copenhagen Design Support Centre, Statutory Section  
Strandvejen 104A, 2nd floor  
DK-2900 Hellerup  
Denmark

Date: **24 June 2010**

This certificate is issued to **DHI Maritime Technology Evaluation Facility at Hundested, Denmark**

### DHI Maritime Technology Evaluation Facility at Hundested, Denmark

The Document(s) listed in paragraph 1 of the appendix have been examined for compliance with:

- Resolution MEPC.174(58), Annex part 2

and are found to comply from quality assurance and quality control aspects subject to the following:

- 1.1. It is required to maintain full and accurate log files in order to demonstrate correct quality measures
- 1.2. The Quality Assurance Project Plan is a project specific document and should as such be subject to review and commenting prior to each project start-up.
- 1.3. This design appraisal document is to be kept together with quality management plan.
- 1.4. Subject certificate is valid until 23 June 2015.

1. The documents listed below have been examined

Drawing No.	Rev.	Title	Status	Date
<b>Date: 23 June 2010</b>	<b>1</b>	<b>Quality Management Plan</b>	<b>B</b>	<b>24 June 2010</b>

2. The documents listed below have been considered together with the submitted documents in the appraisal

Drawing No.	Rev.	Title
<b>11806056</b>	<b>02</b>	<b>Quality Assurance Project Plan</b>

#### Appraisal Status Key

**B** Examined and found to comply with §2.1, Part 2 of the annex of IMO Resolution MEPC 174 (58)

*Martin Schabert*

Martin Schabert  
Statutory Department  
Copenhagen Design Support Centre  
Surveyor to Lloyd's Register EMEA



A member of the Lloyd's Register Group

Lloyd's Register, its affiliates and subsidiaries and their respective officers, employees or agents are, individually and collectively, referred to in this clause as the 'Lloyd's Register Group'. The Lloyd's Register Group assumes no responsibility and shall not be liable to any person for any loss, damage or expense caused by reliance on the information or advice in this document or howsoever provided, unless that person has signed a contract with the relevant Lloyd's Register Group entity for the provision of this information or advice and in that case any responsibility or liability is exclusively on the terms and conditions set out in that contract.

Form 1124 (2005.02)

This is a copy of an electronic document. In the event of any conflict of ambiguity between the copy and the electronic document, which is retained and published by Lloyd's Register, the original electronic and certified version shall always prevail.



## ***A P P E N D I X   B***

### ***Specification of pumps and other equipment at the test facility***

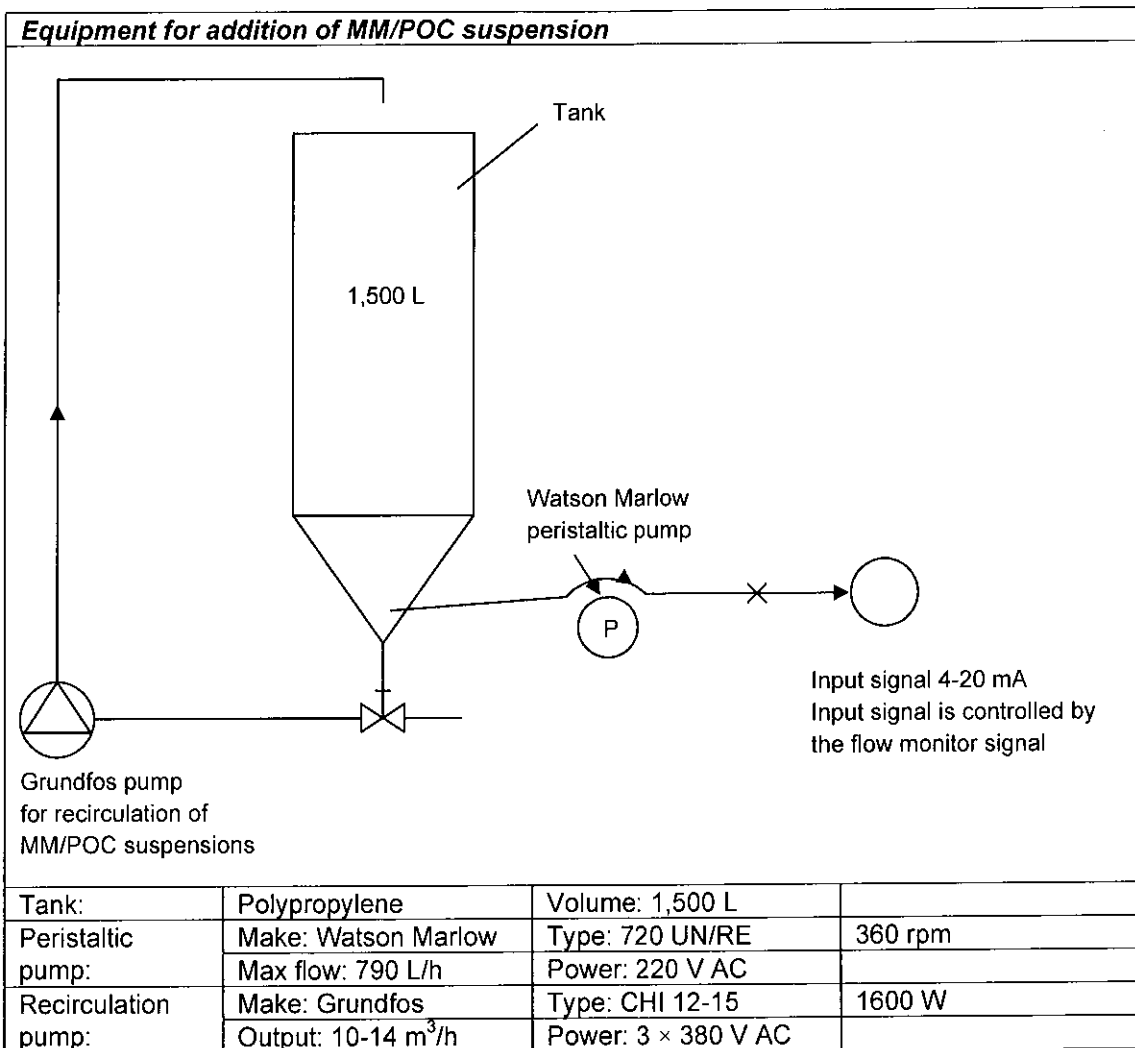


Stirrers			
Make:	LANDIA		
In Tanks A, B & C			
Type:	POP-I	3.0 kW	300 rpm
In Tank D			
Type:	POP-I	5.5 kW	300 rpm

<b>Main pump</b>			
Make:	DESMI		
Type:	NSL 200-330/DOZ-P		
Output:	500 m <sup>3</sup> /h at 25 m water column		
Power:	3 × 380 V AC	55 kW	
Paint:	Inside of pump housing painted with HEMPADUR 35530		
Adjustability:	Adjustable by use of Danfoss VLT FC 202	20 Hz → 50 Hz	

<b>High pressure cleaner</b>			
Make:	Nilfisk ALTO		
Type:	4-55 FAX		
Output:	200 bar	1 m <sup>3</sup> /h	
Temperature:	10-100°C		

<b>Wet&amp;dry vacuum cleaner</b>			
Make:	Nilfisk ALTO		
Type:	ATTIX 965-21 SD XC		
Output:	Vacuum 230 mbar		
Power	380 V AC	2 × 1.1 kW	



Marine water inlet - Submersible pump			
Make:	Grindex		
Type:	Minor Proline	No.: 223963	
Power:	3 × 380 V AC	4.5 kW	
Output:	62 m <sup>3</sup> /h at 37 m water column		

Freshwater collection – Pump			
Make:	HONDA		
Type:	WT 40XK2		
Power:	Petrol	11 kW	
Output:	138 m <sup>3</sup> /h at 29 m water column		

Auxiliary pump for addition of cultures			
Make:	HONDA		
Type:	VB20		
Power:	Petrol	3.5 kW	
Output:	36 m <sup>3</sup> /h at 32 m water column		



## **A P P E N D I X C**

***Description of the BIO UV ballast water management system BIO-SEA  
as given by the client***

## 1. GENERAL DESCRIPTION OF BIO-SEA SYSTEM

BIO-SEA is a system specifically designed for ballast water treatment. It consists of two treatment steps:

- Step 1: **MECHANICAL FILTRATION**. It aims at reducing the amount of total suspended particles, organic or not, present in the sea water.
- Step 2: **ULTRAVIOLET DISINFECTION**. Without any addition of chemicals nor creation of active substances, it inactivates the microorganisms present in the water (bacteria, phytoplankton, zooplankton).

When ballasting, both operations of filtration and UV disinfection are carried out: the objective is to limit the loading of suspended solids and living microorganisms in the ballast tanks.

When deballasting, only the operation of UV disinfection is completed (the filter is bypassed). This allows re-treating the water that stayed in the ship's ballast tanks during the journey, in order to eliminate the possible biological recontamination, to ensure compliance with the IMO standards for discharge of ballast water.

The entire operation of the BIO-SEA system is automated (valves opening and closing, filter cleaning, UV intensity regulation)

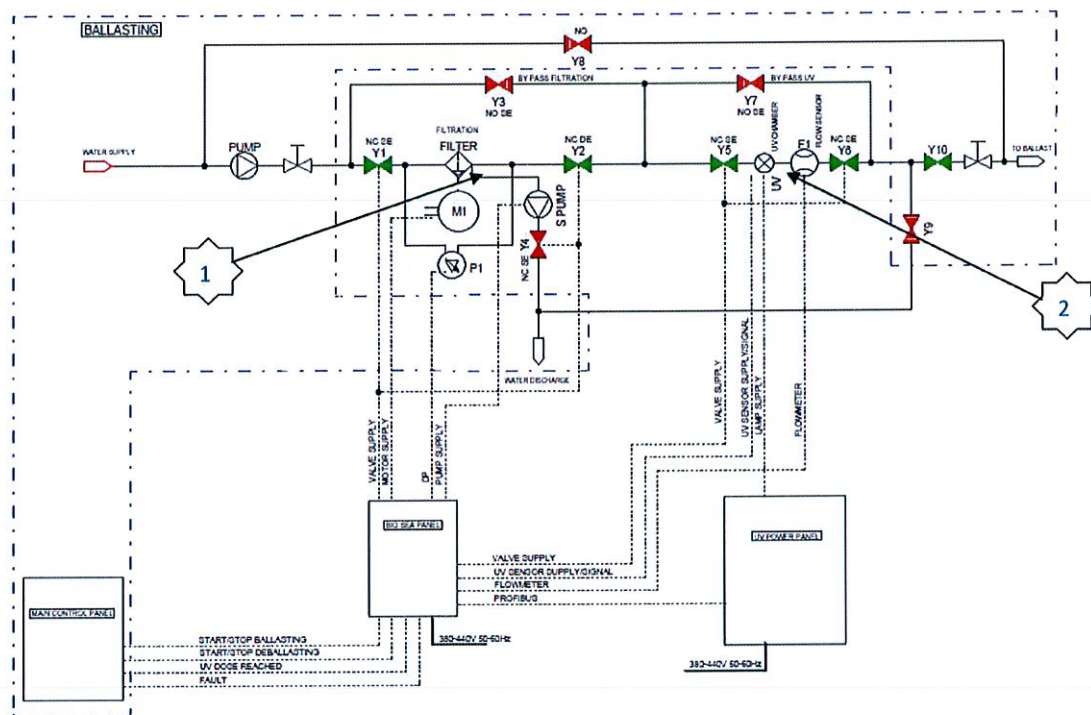


Fig. 1: Piping and controls, during ballasting operation.

## 2. DESCRIPTION OF THE FILTER

The filter presents a cylindrical housing with a filtration screen (mesh) of 40 $\mu$ m. An extended filtering surface allows a good retention rate. The needed surface is dimensioned depending on the flow rate to treat.

Filtration is carried out from inside (entry of dirty water) to outside (clean water).

The clogging level of the filter screen is monitored by a differential pressure switch. When the pressure differential reaches the defined threshold value, indicating filter clogging, a suction device is automatically started to clean the screen.

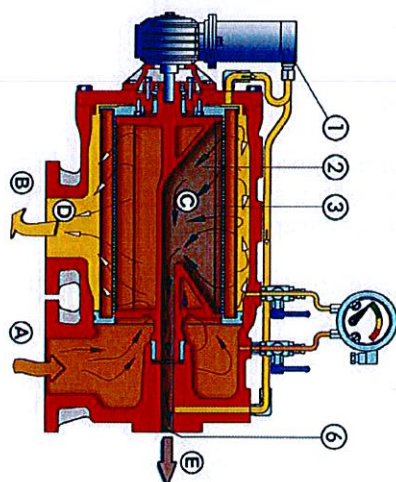
A specific pump boosts the suction process to unstick the clogged solids, and drove them back to the original seawater (harbor) through the discharge pipe, so that the suspended solids and biggest organisms are pumped back into the medium from which they came.

The cleaning cycle of the filter screen does not disrupt the filtration process, allowing no significant variation of the treated flow rate.

At the end of the ballasting operation, the filter is drained and refilled with fresh water.

2 models of filter can be installed within the BIO-SEA treatment system, whose technical characteristics are identical. Only the overall shape and position (dimensions, footprint) of the filter is different.

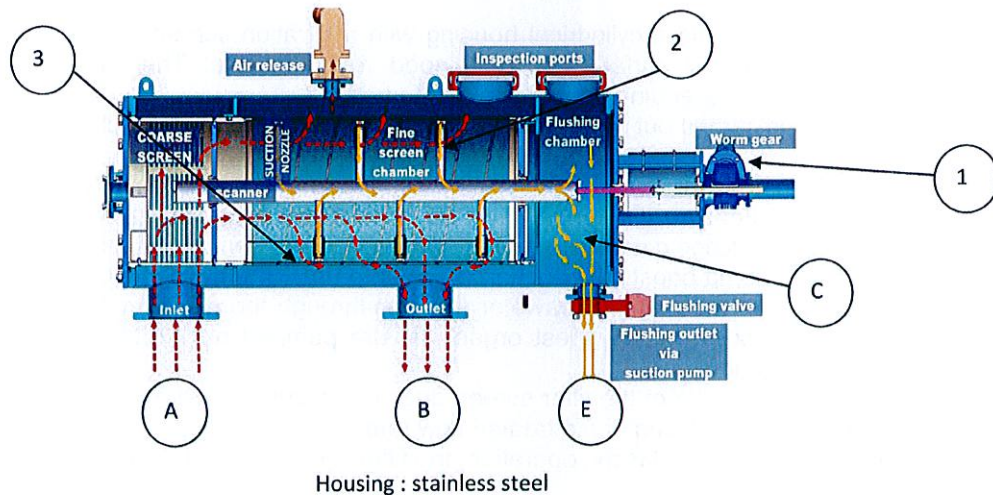
**Fig.2: FILTER 1, in vertical position**



1	Gear and Motor for self-cleaning system
2	Succion nozzle
3	Filtration screen
A	Inlet of untreated seawater
B	Outlet of treated water
C	Backflush water
E	Flush water discharge

Cast Housing (bronze-aluminum)

**Fig.3: FILTER 2, in horizontal position**



1	Gear and Motor for self-cleaning system
2	Suction nozzle
3	Filtration screen
A	Inlet of untreated seawater
B	Outlet of treated water
C	Backflush water
E	Flush water discharge

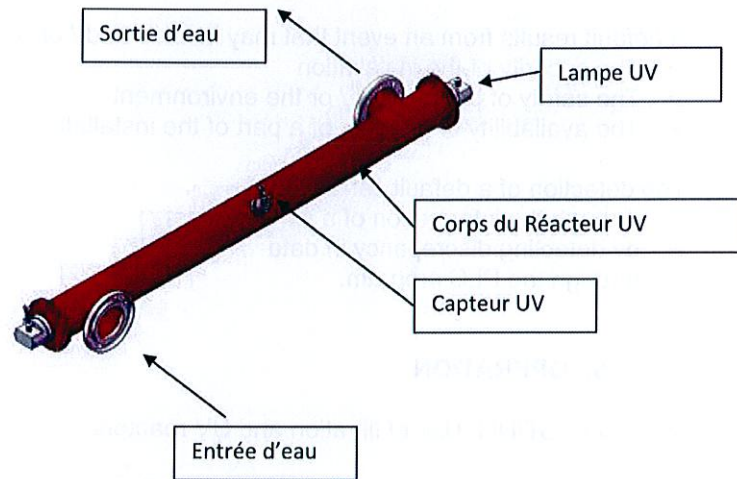
### 3. DESCRIPTION OF UV REACTORS

Designed by CFD (Computational Fluid Dynamic), the UV reactor used in the BIO-SEA system is a tubular-type reactor equipped with a single high power lamp placed in a central position. This design has been optimized considering the quality of seawater to be treated, especially its UV transmittance.

As well, the fluid speeds in the reactor were studied in order to avoid deposits on the quartz sleeves, and thus limit the cleaning and maintenance needs.

The characteristics of the BIO-SEA UV reactors are:

- Reactor equipped with a single medium pressure UV lamp, polychromatic, high intensity.
- Protection of the UV lamp in a quartz sleeve of high purity,
- Lamp driven by electronic ballast, allowing precise management of the UV lamp in order to optimize its regulation, reduce the power consumption and prolong its life
- Monitoring through UV sensor (intensity),
- Modular design that facilitates the installation of UV reactors in parallel, and a better adjustment to the flow that has to be treated.



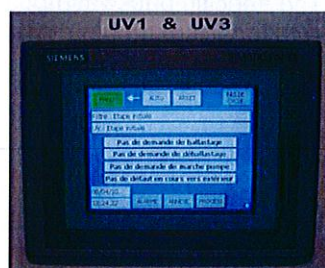
**Fig.4: UV Reactor**

#### 4. DESCRIPTION OF CONTROL / COMMAND

A PLC allows to automatically operate and monitor the BIO-SEA treatment system  
The main functions are:

- Automatic management of "ballasting" and "deballasting" modes
- Monitoring of the operation of the filtration device and UV reactors, thanks to the installation of different sensors:
  - UV sensor, placed on the UV reactor: monitoring that UV intensity is above a target value (expressed in  $\text{w/m}^2$ ), it ensures the good operation of UV disinfection.
  - Temperature sensor ( $^{\circ}\text{C}$ )
  - Flow meter ( $\text{m}^3/\text{h}$ )
  - Differential pressure switch (between filter inlet and outlet) to control clogging cycle (bars).
  - End stroke switch on all automatic valves
- Recording of operations, alarms and measured UV intensity (10,000 records each) to cover a history of 24 months

The user interface is composed of a touch screen, allowing good ergonomics and an easier understanding of the process.



**Fig.5: Image display PLC**

#### DEFAULT MANAGEMENT (ALARMS)

A default results from an event that may involve and / or be originated by:

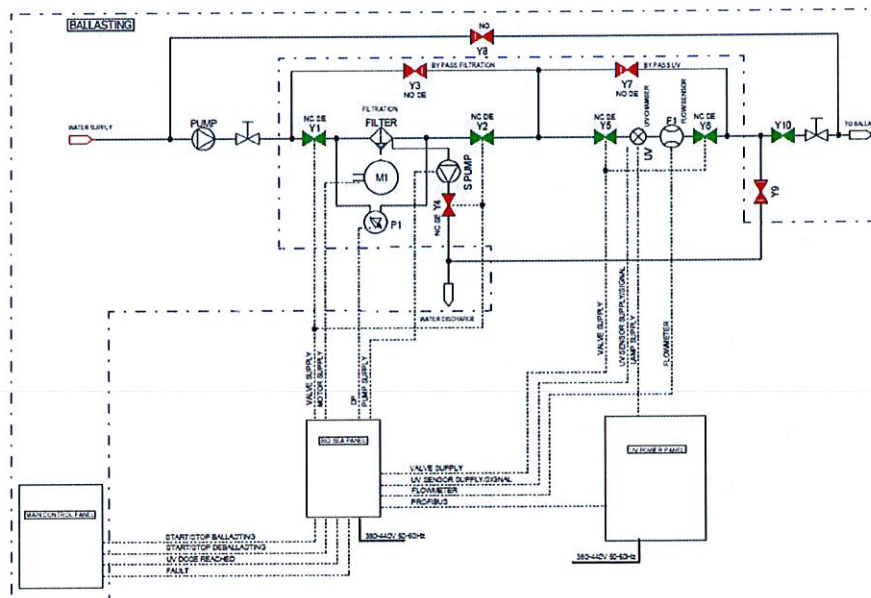
- The security of the installation
- The safety of people and / or the environment
- The availability or integrity of a part of the installation

The detection of a default can be done:

- through the information of a sensor
- by detecting discrepancy in data
- through the PLC program.

## 5. OPERATION

- **BALLASTING:** Use of filtration and UV reactors.

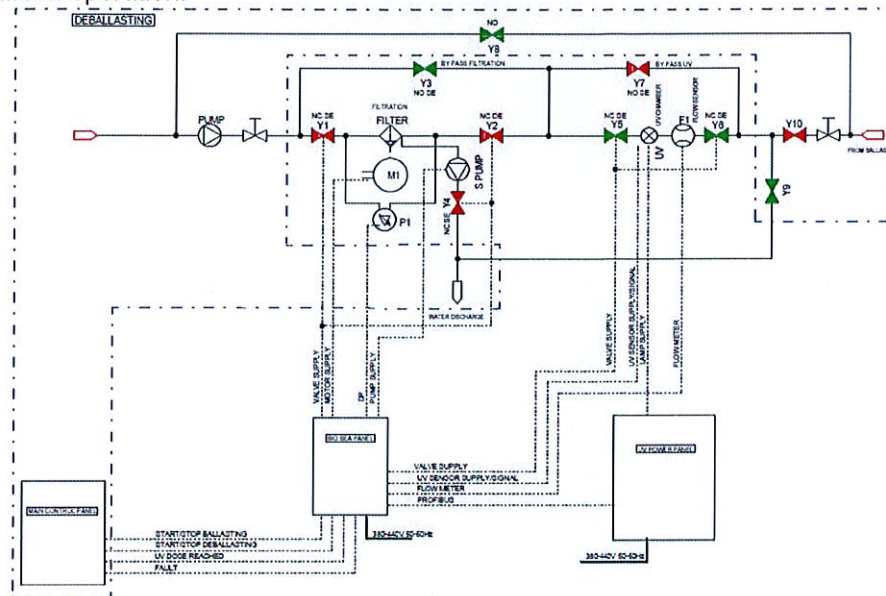


**Fig. 6: Ballasting operation**

When the pressure differential indicates clogging of the filter, the self-cleaning cycle is automatically launched. The specific backwash pump ensures the discharge of the filtrate, without loss of flow rate for filling the ballast tanks.



- **DEBALLASTING:** The filter is automatically by-passed and only the UV reactors are in operation.



**Fig. 7: Debballasting operation**

- At the end of operation (either ballasting or deballasting) a total purge of the system is performed, flushing and filling the filter and reactors with fresh water.

#### **MODES:**

- **AUTO:** automated operation of the system. This mode manages control systems, regulations and decisions on cycle defects, including the system security procedures.
- **MANUAL:** this mode allows operating the system (mechanisms) manually, to conduct basic operations when:
  - the automatic mode is no longer available
  - a mechanism or element must be positioned in a defined state.
- **MAINTENANCE:** this mode allows operating all mechanisms during repairing or maintenance. No security is provided in this mode. Recording remains the responsibility of the user.
- **STOP:** This mode triggers a shutdown of the software / system. Only the monitoring process is possible.
- **EMERGENCY STOP:** This emergency shutdown must be activated by an operator by pushing the "emergency stop" button. This mode puts the relevant mechanisms in a safe condition (PLC, remote mechanisms). Management of inputs (digital, analog) remains intact.

#### **ACCESS TO THE SYSTEM:**

To validate or block certain actions / operations, entering passwords may be necessary ; these validations are under two levels of responsibility:

- Operator Mode: operation of the system, actions on cycles
- Administrator Mode: same as operator + maintenance mode + settings.



## ***A P P E N D I X   D***

### ***Overview of SOPs***



SUBJECT/SUBSUBJECT	SOP NO.
ANALYTICAL METHOD ZOOPLANKTON ANALYSIS	30/1700:04
ANALYTICAL METHOD MICROSCOPIC ENUMERATION AND IDENTIFICATION OF MICROALGAE (LUGOL AND CMFDA/FDA)	30/1701:02
ANALYTICAL METHOD DETERMINING PRIMARY PRODUCTION OF MICROALGAE	30/1702:03
ANALYTICAL METHOD DETERMINING DIVERSITY OF MICROALGAE COMMUNITIES BY HPLC ANALYSIS OF PIGMENTS	30/1703:03
ANALYTICAL METHOD DETERMINATION OF VIABLE ALGAE BY MPN	30/1704:02
MICROBIOLOGICAL TESTS DETERMINATION OF TOTAL NUMBER OF BACTERIA BY EPIFLUORESCENCE MICROSCOPY	30/1705:03
MICROBIOLOGICAL TESTS DETERMINATION OF HETEROTROPHIC PLATE COUNT	30/1706:03
MICROBIOLOGICAL TESTS DETERMINATION OF <i>VIBRIO CHOLERAE</i> IN WATER	30/1707:02
MICROBIOLOGICAL TESTS DETERMINATION OF TOTAL COLIFORM, <i>E. COLI</i> AND ENTEROCOCCI Colilert*-18 AND Enterolert-E	30/1708:02
HARVESTING, CULTURING AND ADDITION OF ORGANISMS	30/1734:03
COLLECTION OF SEAWATER	30/1735:02
COLLECTION OF FRESH WATER	30/1736:02
CHEMICAL CRITERIA FOR TEST WATER ADDITION OF DOC, POC AND MM	30/1737:02
SAMPLING BIOLOGICAL AND WATER QUALITY PARAMETERS	30/1738:02
SAMPLING WET TEST	30/1739:02
STATISTICS STATISTICAL SURVEILLANCE OF BIOLOGICAL DATA OBTAINED AT TESTS OF BWMSs	30/1760:01
LABELLING SAMPLES COLLECTED AT TEST SITE	30/1761:01
OPERATION OF THE DHI MTEF	30/1762:02
CLEANING RETENTION TANKS; PIPINGS AND OTHER EQUIPMENT AT TEST SITE	30/1763:02
MEASUREMENT METHOD ON-LINE MONITORING OF PRESSURE, TEMPERATURE AND FLOW RATES AT TEST SITE	30/1764:01
MEASUREMENT METHOD TURBIDITY	30/1766:03
HEALTH AND SAFETY ENSURING WORKER HEALTH AND SAFETY AT TEST SITE	30/1767:02
MEASUREMENT METHOD DETERMINATION OF TSS	30/1768:02
MEASUREMENT METHOD DETERMINATION OF DOC AND POC	30/1769:02

## AMENDMENT

QAPP DOCUMENT TITLE AND DATE:

Quality Assurance Project Plan

Land-based Test of BIO-SEA Ballast Water Management System (1<sup>st</sup> of September 2011)

AMENDMENT NUMBER:

1

DATE OF AMENDMENT:

10<sup>th</sup> of October 2011

AMENDMENT CONTENTS:

The proposed test schedule is changed from:

Month	September			October		November		
Week No.	35	36	38	40	42	44	45	46
Test cycle No.	X (1) X+1	2 3	4 5	Sup1	6 7	8 9	10	Sup2

To:

Month	September			October			November
Week No.	35	36	38	40	41	42	44
Test cycle No.	X (1) X+1	2 3	4 5	Sup1 6	7	8 9	10 Sup2

REASON FOR AMENDMENT:

The test schedule has been updated due to logistic reasons

IMPACT OF AMMENDMENT:

The only impact of this amendment is early finalization of the land-based testing.

PREVENTATIVE ACTION:

Not relevant.



ORIGINATED BY:

DHI

SIGNED BY:

Morten Bjergstrøm  
Project manager

10<sup>th</sup> of October 2011  
DATE

Copy to be sent to the client, the Certification Body and the DHI Quality Assurance Unit.

## AMENDMENT

QAPP DOCUMENT TITLE AND DATE:

Quality Assurance Project Plan

Land-based Test of BIO-SEA Ballast Water Management System (1<sup>st</sup> of September 2011)

AMENDMENT NUMBER:

2

DATE OF AMENDMENT:

14<sup>th</sup> of October 2011

AMENDMENT CONTENTS:

The proposed test schedule is changed from:

Month	September			October			November
Week No.	35	36	38	40	41	42	44
Test cycle No.	X (1) X+1	2 3	4 5	Sup1 6	7	8 9	10 Sup2

To:

Month	September			October			
Week No.	35	36	38	40	41	42	43
Test cycle No.	X (1) X+1	2 3	4 5	Sup1 6	7	8 Sup2	9 10

REASON FOR AMENDMENT:

The test schedule has been updated due to logistic reasons

IMPACT OF AMMENDMENT:

The only impact of this amendment is early finalization of the land-based testing.

PREVENTATIVE ACTION:

Not relevant.



ORIGINATED BY:

DHI

SIGNED BY:

Morten Bjergstrøm  
Project manager

14<sup>th</sup> of October 2011  
DATE

Copy to be sent to the client, the Certification Body and the DHI Quality Assurance Unit.



## **A P P E N D I X   B**

Data logging from the land-based testing

Table B-1 Test cycle B-1

Description	Data	Comments
Client treatment system	BIO-SEA	
Salinity (PSU)	17.1	
Retention tank no.	C	
Test cycle	B-1	
System cleaned before inlet	Yes	
Date and time inlet start	2011.09.01 09:56	
Date and time inlet stop	2011.09.01 10:22	
Flow during inlet (average)	506 m <sup>3</sup> /h	
Power consumption during inlet	138 kW	Provided by BIO-UV
Treated volume during inlet	212 m <sup>3</sup>	
App. volume used for treatment	223 m <sup>3</sup>	
System cleaned before discharge	Yes	
Date and time discharge start	2011.09.06 08:36	
Date and time discharge stop	2011.09.06 09:00	
Flow during discharge (average)	507 m <sup>3</sup> /h	
Power consumption during discharge	138 kW	Provided by BIO-UV
Treated volume during discharge	212 m <sup>3</sup>	
Use of cultured organisms	Yes	<i>Artemia</i> and <i>Tetraselmis</i>
Filter	Filtrex	
UV units	6	

Table B-2 Test cycle B-1

On-line measurements	DO sat. (%)	pH	Salinity (PSU)	Temperature (C°)	Turbidity (NTU)
Control inlet (T0)	95.81±0.04	8.07±0.00	16.72±1.60	16.05±0.01	29.38±0.57
BIO-SEA inlet (T0)	96.03±0.23	8.06±0.00	17.08±0.00	16.04±0.01	31.47±3.75
BIO-SEA 1 <sup>st</sup> treatment (T0)	97.19±4.12	8.00±0.01	16.71±1.77	16.25±0.04	33.21±2.03
BIO-SEA inlet (T5)	71.86±1.60	7.69±0.01	17.09±0.02	17.25±0.00	20.83±1.74
BIO-SEA 2 <sup>nd</sup> treatment (T5)	74.78±7.62	7.63±0.02	17.00±0.37	17.44±0.09	21.26±1.45
Control discharge (T5)	74.04±3.45	7.71±0.00	17.09±0.02	17.73±0.03	25.56±1.60

Table B-3 Test cycle B-2

Description	Data	Comments
Client treatment system	BIO-SEA	
Salinity (PSU)	17.1	
Retention tank no.	B	
Test cycle	B-2	
System cleaned before inlet	Yes	
Date and time inlet start	2011.09.01 11:47	
Date and time inlet stop	2011.09.01 12:13	
Flow during inlet (average)	511 m <sup>3</sup> /h	
Power consumption during inlet	115 kW	Provided by BIO-UV
Treated volume during inlet	205 m <sup>3</sup>	
App. volume used for treatment	232 m <sup>3</sup>	
System cleaned before discharge	Yes	
Date and time discharge start	2011.09.01 10:53	
Date and time discharge stop	2011.09.01 11:16	
Flow during discharge (average)	507 m <sup>3</sup> /h	
Power consumption during discharge	115 kW	Provided by BIO-UV
Treated volume during discharge	205 m <sup>3</sup>	
Use of cultured organisms	Yes	<i>Artemia and Tetraselmis</i>
Filter	Filtrex	
UV units	5	

Table B-4 Test cycle B-2

On-line measurements	DO sat. (%)	pH	Salinity	Temperature (C°)	Turbidity (NTU)
Control inlet (T0)	95.81±0.04	8.07±0.00	16.72±1.60	16.05±0.01	29.38±0.57
BIO-SEA inlet (T5)	96.80±0.68	8.07±0.00	17.08±0.01	16.20±0.03	30.53±1.28
BIO-SEA 1 <sup>st</sup> treatment (T0)	96.64±1.35	8.01±0.01	17.07±0.13	16.62±0.73	33.39±1.25
BIO-SEA inlet (T5)	77.18±1.22	7.78±0.00	17.08±0.01	17.68±0.01	23.44±5.42
BIO-SEA 2 <sup>nd</sup> treatment (T5)	78.86±4.13	7.72±0.00	17.06±0.01	17.86±0.01	22.23±1.08
Control discharge (T5)	74.04±3.45	7.71±0.00	17.09±0.02	17.73±0.03	25.56±1.60

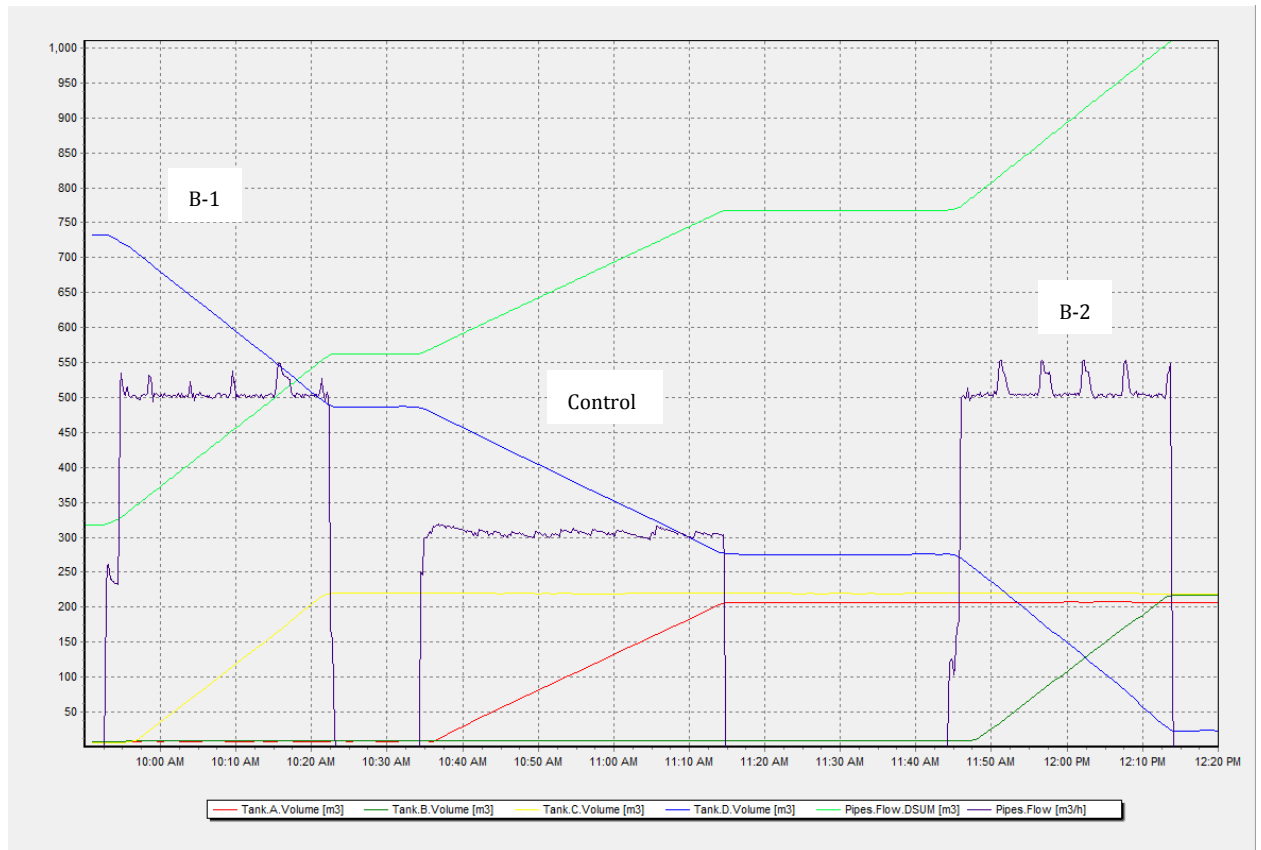


Figure B-1 BIO-SEA test cycles B-1 and B-2 inlet

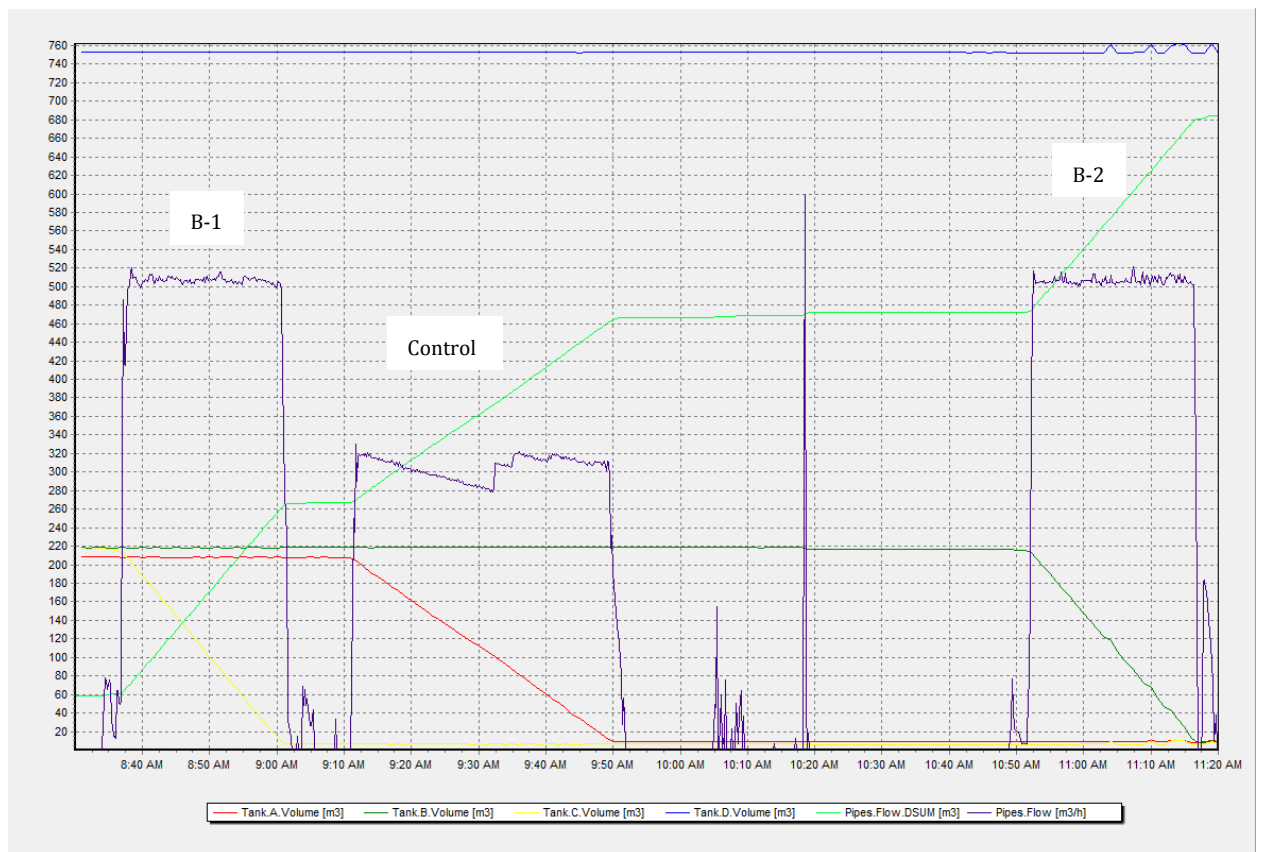


Figure B-2 BIO-SEA test cycles B-1 and B-2 discharge

Table B-5 Test cycle B-3

Description	Data	Comments
Client treatment system	BIO-SEA	
Salinity (PSU)	17.2	
Retention tank no.	C	
Test cycle	B-3	
System cleaned before inlet	Yes	
Date and time inlet start	2011.09.08 08:52	
Date and time inlet stop	2011.09.08 09:17	
Flow during inlet (average)	504 m <sup>3</sup> /h	
Power consumption during inlet	115 kW	Provided by BIO-UV
Treated volume during inlet	210 m <sup>3</sup>	
App. volume used for treatment	213 m <sup>3</sup>	
System cleaned before discharge	Yes	
Date and time discharge start	2011.09.13 08:46	
Date and time discharge stop	2011.09.13 09:08	
Flow during discharge (average)	511 m <sup>3</sup> /h	
Power consumption during discharge	115 kW	Provided by BIO-UV
Treated volume during discharge	210 m <sup>3</sup>	
Use of cultured organisms	Yes	<i>Artemia and Tetraselmis</i>
Filter	Filtrex	
UV units	5	

Table B-6 Test cycle B-3

On-line measurements	DO sat. (%)	pH	Salinity	Temperature (C°)	Turbidity (NTU)
Control inlet (T0)	98.94±0.12	8.18±0.00	17.24±0.01	15.67±0.00	30.85±0.26
BIO-SEA inlet (T0)	99.77±0.43	8.18±0.00	17.24±0.00	15.72±0.01	32.77±1.06
BIO-SEA 1 <sup>st</sup> treatment (T0)	99.71±1.12	8.10±0.00	17.22±0.03	15.89±0.02	32.02±0.98
BIO-SEA inlet (T5)	70.75±2.51	7.71±0.00	17.23±0.02	16.96±0.01	18.51±4.10
BIO-SEA 2 <sup>nd</sup> treatment (T5)	73.14±2.43	7.65±0.03	17.19±0.01	17.13±0.01	18.12±0.57
Control discharge (T5)	75.20±1.03	7.80±0.00	17.23±0.01	17.37±0.02	24.28±1.23

Table B-7 Test cycle B-4

Description	Data	Comments
Client treatment system	BIO-SEA	
Salinity (PSU)	17.2	
Retention tank no.	B	
Test cycle	B-4	
System cleaned before inlet	Yes	
Date and time inlet start	2011.09.08 11:03	
Date and time inlet stop	2011.09.08 11:28	
Flow during inlet (average)	504 m <sup>3</sup> /h	
Power consumption during inlet	93 kW	Provided by BIO-UV
Treated volume during inlet	203 m <sup>3</sup>	
App. volume used for treatment	215 m <sup>3</sup>	
System cleaned before discharge	Yes	
Date and time discharge start	2011.09.13 11:22	
Date and time discharge stop	2011.09.13 11:43	
Flow during discharge (average)	507 m <sup>3</sup>	
Power consumption during discharge	92 kW	Provided by BIO-UV
Treated volume during discharge	203 m <sup>3</sup>	
Use of cultured organisms	Yes	<i>Artemia and Tetraselmis</i>
Filter	Filtrex	
UV units	4	

Table B-8 Test cycle B-4

On-line measurements	DO sat. (%)	pH	Salinity	Temperature (C°)	Turbidity (NTU)
Control inlet (T0)	98.94±0.12	8.18±0.00	17.24±0.01	15.67±0.00	30.85±0.26
BIO-SEA inlet (T0)	99.41±0.13	8.19±0.00	17.24±0.00	15.73±0.01	31.91±0.42
BIO-SEA 1 <sup>st</sup> treatment (T0)	99.58±0.29	8.11±0.00	17.22±0.01	15.86±0.01	32.16±0.54
BIO-SEA inlet (T5)	71.55±0.78	7.78±0.00	17.24±0.01	17.23±0.01	18.14±2.65
BIO-SEA 2 <sup>nd</sup> treatment (T5)	72.88±0.90	7.72±0.00	17.17±0.01	17.38±0.01	19.04±0.66
Control discharge (T5)	75.20±1.03	7.80±0.00	17.23±0.01	17.37±0.02	24.28±1.23

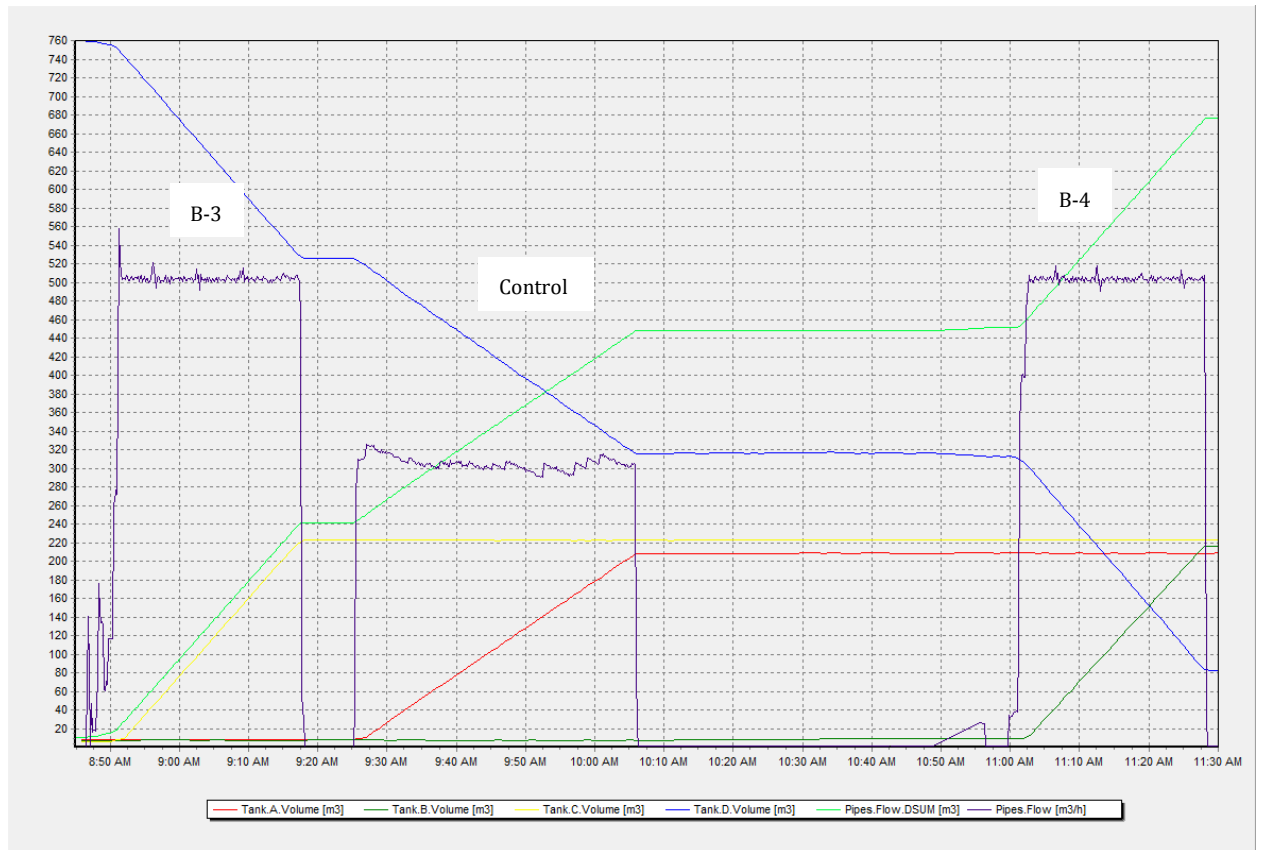


Figure B-3 BIO-SEA test cycles B-3 and B-4 inlet

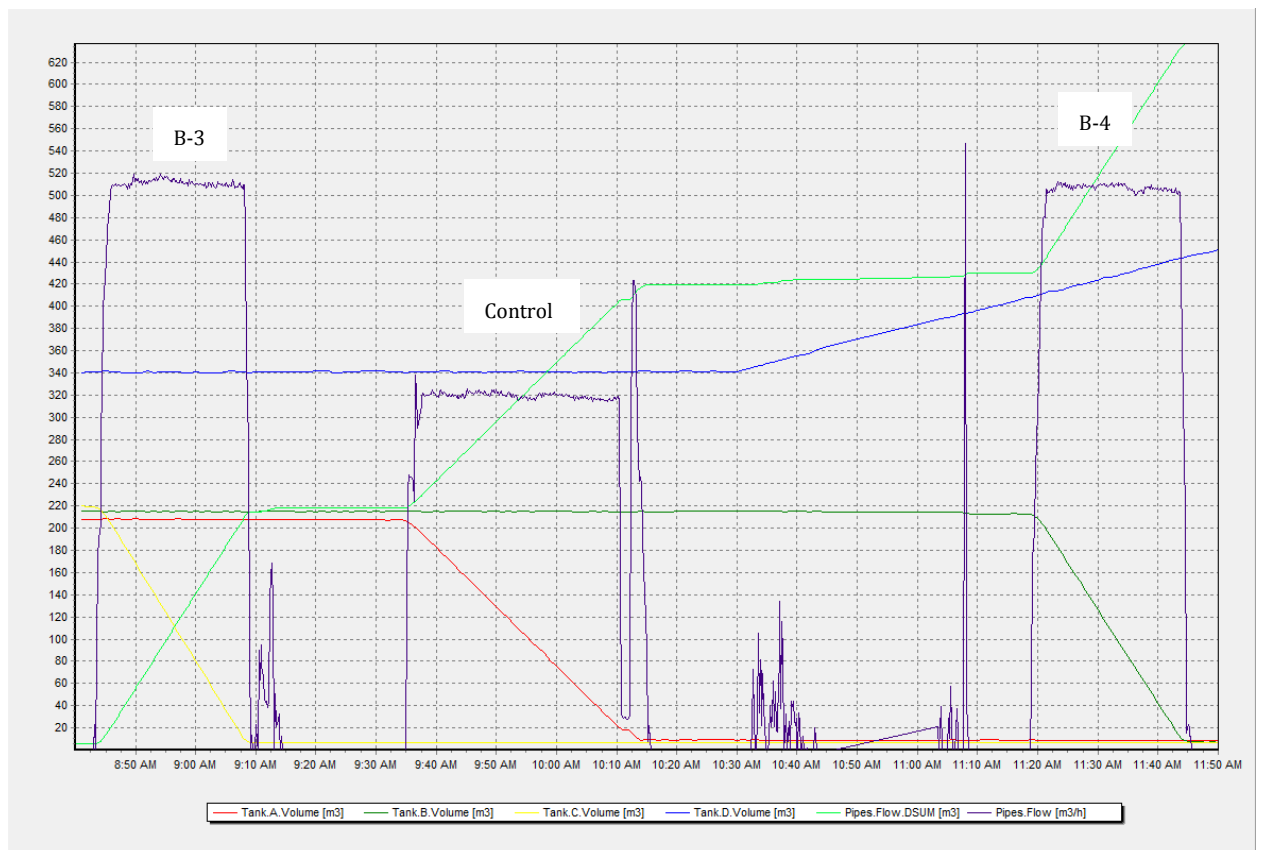


Figure B-4 BIO-SEA test cycles B-3 and B-4 discharge

Table B-9 Test cycle M-1

Description	Data	Comments
Client treatment system	BIO-SEA	
Salinity (PSU)	35.9	
Retention tank no.	C	
Test cycle	M-1	
System cleaned before inlet	Yes	
Date and time inlet start	2011.09.22 09:18	
Date and time inlet stop	2011.09.22 09:44	
Flow during inlet (average)	507 m <sup>3</sup> /h	
Power consumption during inlet	76 kW	Provided by BIO-UV
Treated volume during inlet	217 m <sup>3</sup>	
App. volume used for treatment	226 m <sup>3</sup>	
System cleaned before discharge	Yes	
Date and time discharge start	2011.09.27 08:23	
Date and time discharge stop	2011.09.27 08:46	
Flow during discharge (average)	506 m <sup>3</sup> /h	
Power consumption during discharge	76 kW	Provided by BIO-UV
Treated volume during discharge	217 m <sup>3</sup>	
Use of cultured organisms	Yes	<i>Artemia and Tetraselmis</i>
Filter	Filtrex	
UV units	5	

Table B-10 Test cycle M-1

On-line measurements	DO sat. (%)	pH	Salinity	Temperature (C°)	Turbidity (NTU)
Control inlet (T0)	106.28±0.07	7.98±0.00	35.93±0.01	14.60±0.01	3.66±0.20
BIO-SEA inlet (T0)	106.68±0.31	7.96±0.01	35.94±0.00	14.62±0.01	5.29±1.42
BIO-SEA 1 <sup>st</sup> treatment (T0)	107.82±3.04	7.90±0.01	35.76±0.35	14.74±0.02	5.33±0.40
BIO-SEA inlet (T5)	76.20±1.40	7.58±0.01	35.94±0.02	15.46±0.01	2.55±2.01
BIO-SEA 2 <sup>nd</sup> treatment (T5)	77.45±1.77	7.49±0.03	35.85±0.07	15.59±0.01	2.81±1.03
Control discharge (T5)	77.69±0.59	7.59±0.01	35.93±0.09	16.15±0.01	1.43±0.46

Table B-11 Test cycle M-2

Description	Data	Comments
Client treatment system	BIO-SEA	
Salinity (PSU)	35.9	
Retention tank no.	B	
Test cycle	M-2	
System cleaned before inlet	Yes	
Date and time inlet start	2011.09.22 11:19	
Date and time inlet stop	2011.09.22 11:44	
Flow during inlet (average)	507 m <sup>3</sup> /h	
Power consumption during inlet	76 kW	Provided by BIO-UV
Treated volume during inlet	208 m <sup>3</sup>	
App. volume used for treatment	219 m <sup>3</sup>	
System cleaned before discharge	Yes	
Date and time discharge start	2011.09.27 10:55	
Date and time discharge stop	2011.09.27 11:16	
Flow during discharge (average)	504 m <sup>3</sup> /h	
Power consumption during discharge	76 kW	Provided by BIO-UV
Treated volume during discharge	208 m <sup>3</sup>	
Use of cultured organisms	Yes	<i>Artemia and Tetraselmis</i>
Filter	Filtrex	
UV units	5	

Table B-12 Test cycle M-2

On-line measurements	DO sat. (%)	pH	Salinity	Temperature (C°)	Turbidity (NTU)
Control inlet (T0)	106.28±0.07	7.98±0.00	35.93±0.01	14.60±0.01	3.66±0.20
BIO-SEA inlet (T0)	106.04±0.19	7.99±0.00	35.93±0.00	14.71±0.01	6.68±1.66
BIO-SEA 1 <sup>st</sup> treatment (T0)	106.81±0.22	7.92±0.01	35.83±0.01	14.85±0.01	5.62±0.58
BIO-SEA inlet (T5)	82.60±0.40	7.69±0.00	35.96±0.01	15.91±0.00	2.48±0.76
BIO-SEA 2 <sup>nd</sup> treatment (T5)	83.97±0.71	7.63±0.00	35.87±0.00	16.03±0.00	2.89±0.42
Control discharge (T5)	77.69±0.59	7.59±0.01	35.93±0.09	16.15±0.01	1.43±0.46

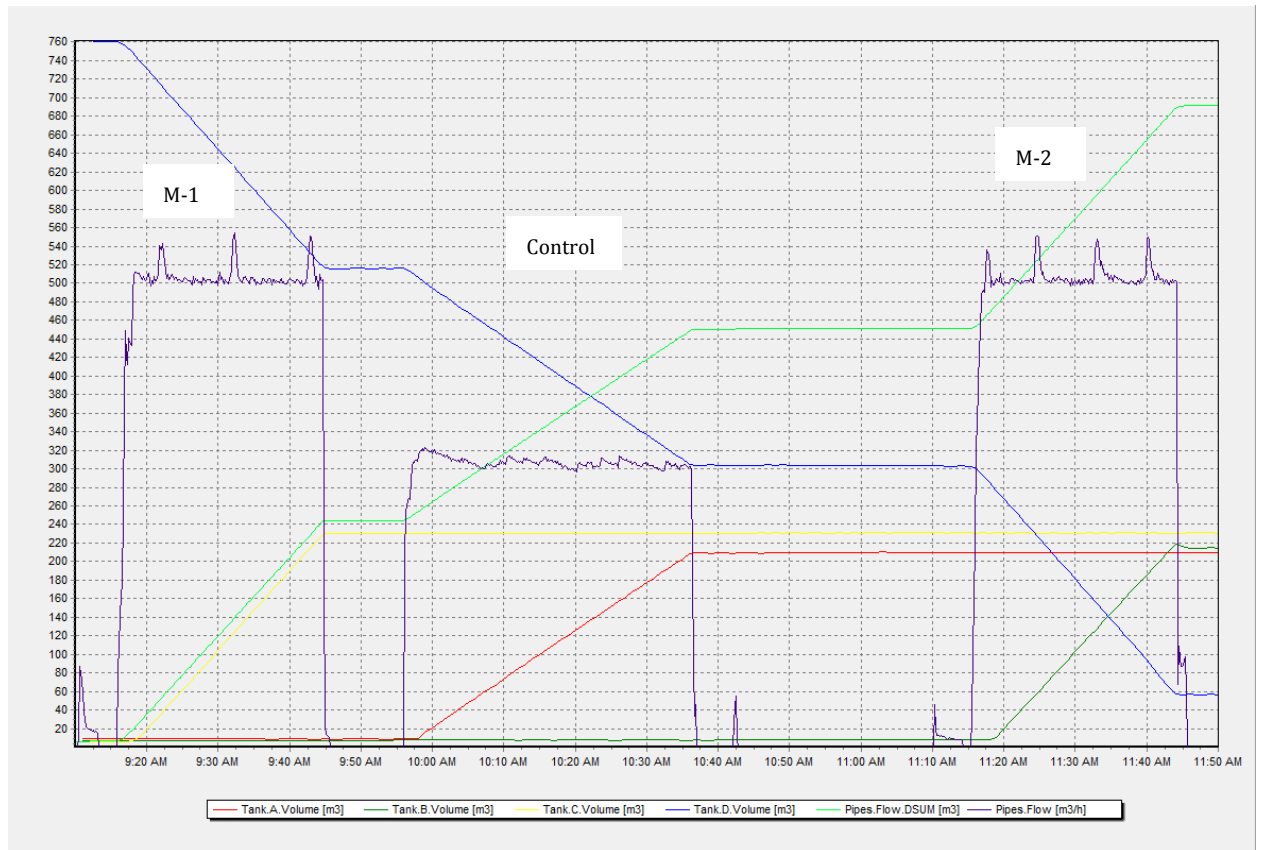


Figure B-5 BIO-SEA test cycles M-1 and M-2 inlet



Figure B-6 BIO-SEA test cycles M-1 and M-2 discharge

Table B-13 Test cycle B-5

Description	Data	Comments
Client treatment system	BIO-SEA	
Salinity (PSU)	17.4	
Retention tank no.	B	
Test cycle	B-5	
System cleaned before inlet	Yes	
Date and time inlet start	2011.10.06 10:19	
Date and time inlet stop	2011.10.06 10:45	
Flow during inlet (average)	514 m <sup>3</sup> /h	
Power consumption during inlet	115 kW	Provided by BIO-UV
Treated volume during inlet	208 m <sup>3</sup>	
App. volume used for treatment	229 m <sup>3</sup>	
System cleaned before discharge	Yes	
Date and time discharge start	2011.10.11 10:25	
Date and time discharge stop	2011.10.11 10:46	
Flow during discharge (average)	506 m <sup>3</sup> /h	
Power consumption during discharge	115 kW	Provided by BIO-UV
Treated volume during discharge	208 m <sup>3</sup>	
Use of cultured organisms	Yes	<i>Artemia and Tetraselmis</i>
Filter	Filtrex	
UV units	5	

Table B-14 Test cycle B-5

On-line measurements	DO sat. (%)	pH	Salinity	Temperature (C°)	Turbidity (NTU)
Control inlet (T0)	98.59±0.07	8.12±0.00	17.41±0.00	14.83±0.00	38.16±0.78
BIO-SEA inlet (T0)	98.80±0.13	8.13±0.00	17.40±0.00	14.89±0.01	37.58±0.68
BIO-SEA 1 <sup>st</sup> treatment (T0)	99.03±0.33	8.07±0.00	17.39±0.00	15.06±0.01	36.71±1.73
BIO-SEA inlet (T5)	83.33±0.63	8.00±0.00	17.40±0.00	12.58±0.00	28.06±2.07
BIO-SEA 2 <sup>nd</sup> treatment (T5)	85.08±2.37	7.93±0.00	17.39±0.02	12.74±0.13	28.08±1.29
Control discharge (T5)	81.47±1.13	7.95±0.00	17.41±0.01	12.57±0.03	29.36±1.73

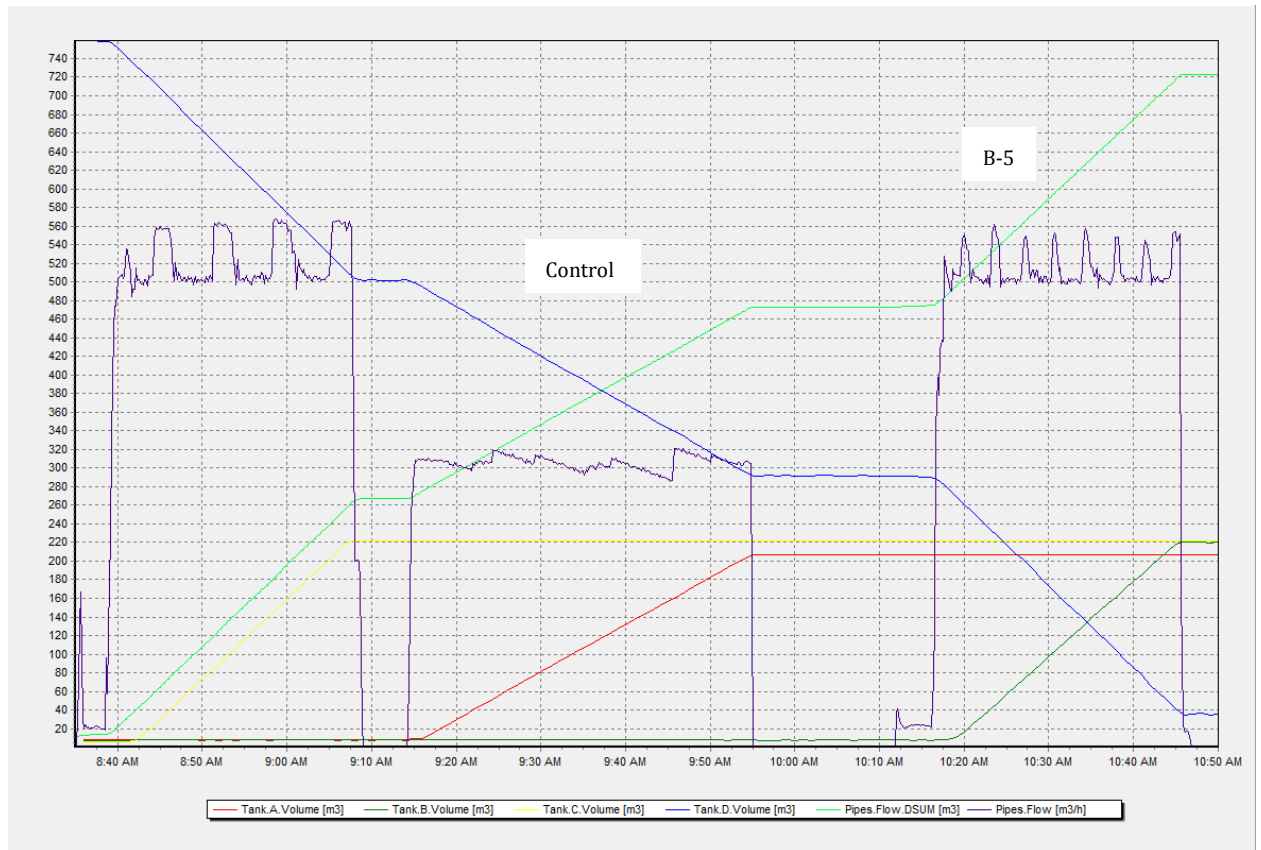


Figure B-7 BIO-SEA test cycles B-5 inlet

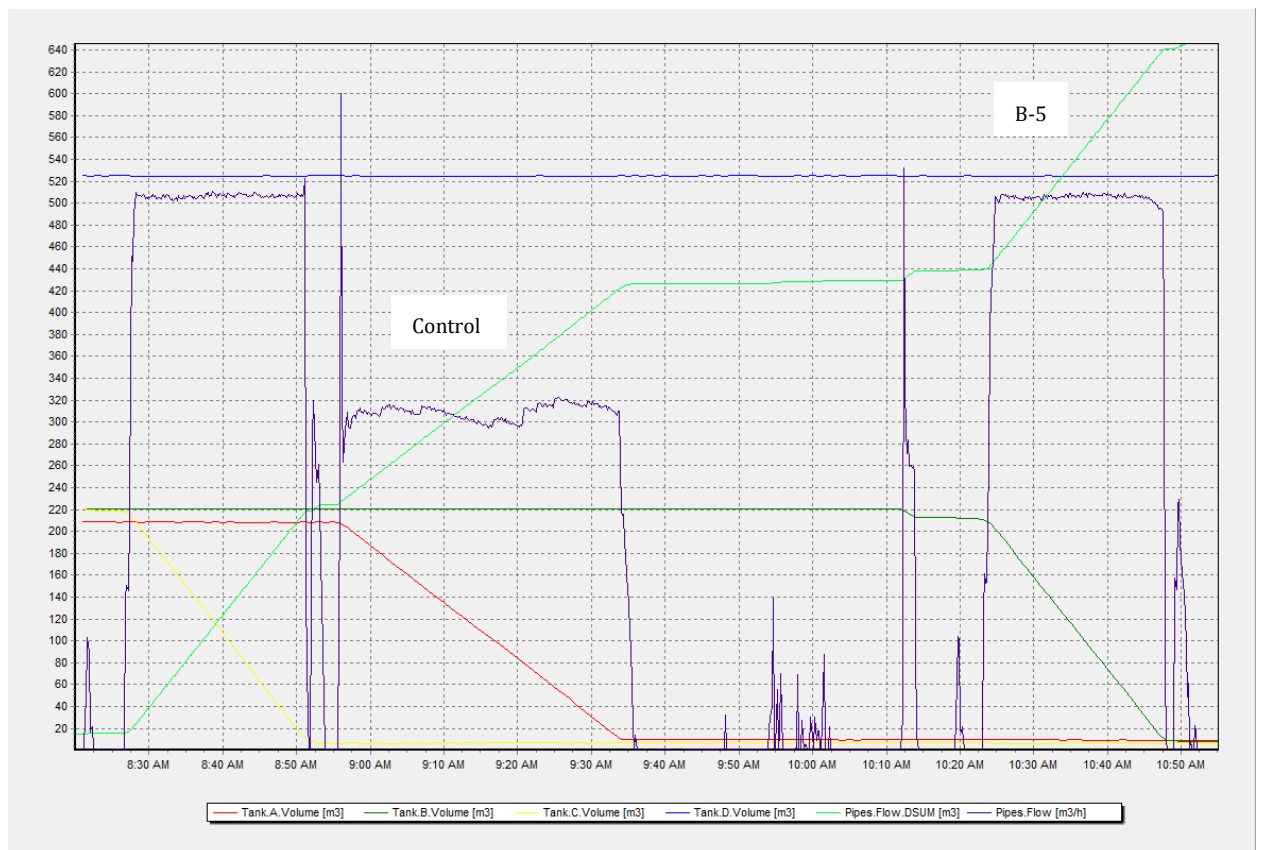


Figure B-8 BIO-SEA test cycles B-5 discharge

Table B-15 Test cycle B-6

Description	Data	Comments
Client treatment system	BIO-SEA	
Salinity (PSU)	18.0	
Retention tank no.	C	
Test cycle	B-6	
System cleaned before inlet	Yes	
Date and time inlet start	2011.10.13 09:12	
Date and time inlet stop	2011.10.13 09:38	
Flow during inlet (average)	514 m <sup>3</sup> /h	
Power consumption during inlet	115 kW	Provided by BIO-UV
Treated volume during inlet	221 m <sup>3</sup>	
App. volume used for treatment	228 m <sup>3</sup>	
System cleaned before discharge	Yes	
Date and time discharge start	2011.10.18 08:09	
Date and time discharge stop	2011.10.18 08:33	
Flow during discharge (average)	507 m <sup>3</sup> /h	
Power consumption during discharge	115 kW	Provided by BIO-UV
Treated volume during discharge	221 m <sup>3</sup>	
Use of cultured organisms	Yes	<i>Artemia and Tetraselmis</i>
Filter	Filtrex	
UV units	5	

Table B-16 Test cycle B-6

On-line measurements	DO sat. (%)	pH	Salinity	Temperature (C°)	Turbidity (NTU)
Control inlet (T0)	98.13±0.51	8.09±0.00	18.04±0.00	11.14±0.08	33.13±1.06
BIO-SEA inlet (T0)	98.07±0.13	8.07±0.01	18.04±0.00	11.21±0.01	33.26±0.58
BIO-SEA 1 <sup>st</sup> treatment (T0)	98.46±0.62	8.00±0.01	18.03±0.00	11.37±0.02	34.46±0.39
BIO-SEA inlet (T5)	86.26±1.21	7.80±0.01	18.03±0.02	10.88±0.00	31.56±7.73
BIO-SEA 2 <sup>nd</sup> treatment (T5)	87.76±3.15	7.74±0.03	17.96±0.03	11.05±0.03	27.89±1.33
Control discharge (T5)	80.59±2.78	7.72±0.02	18.04±0.02	11.68±0.20	29.83±1.85

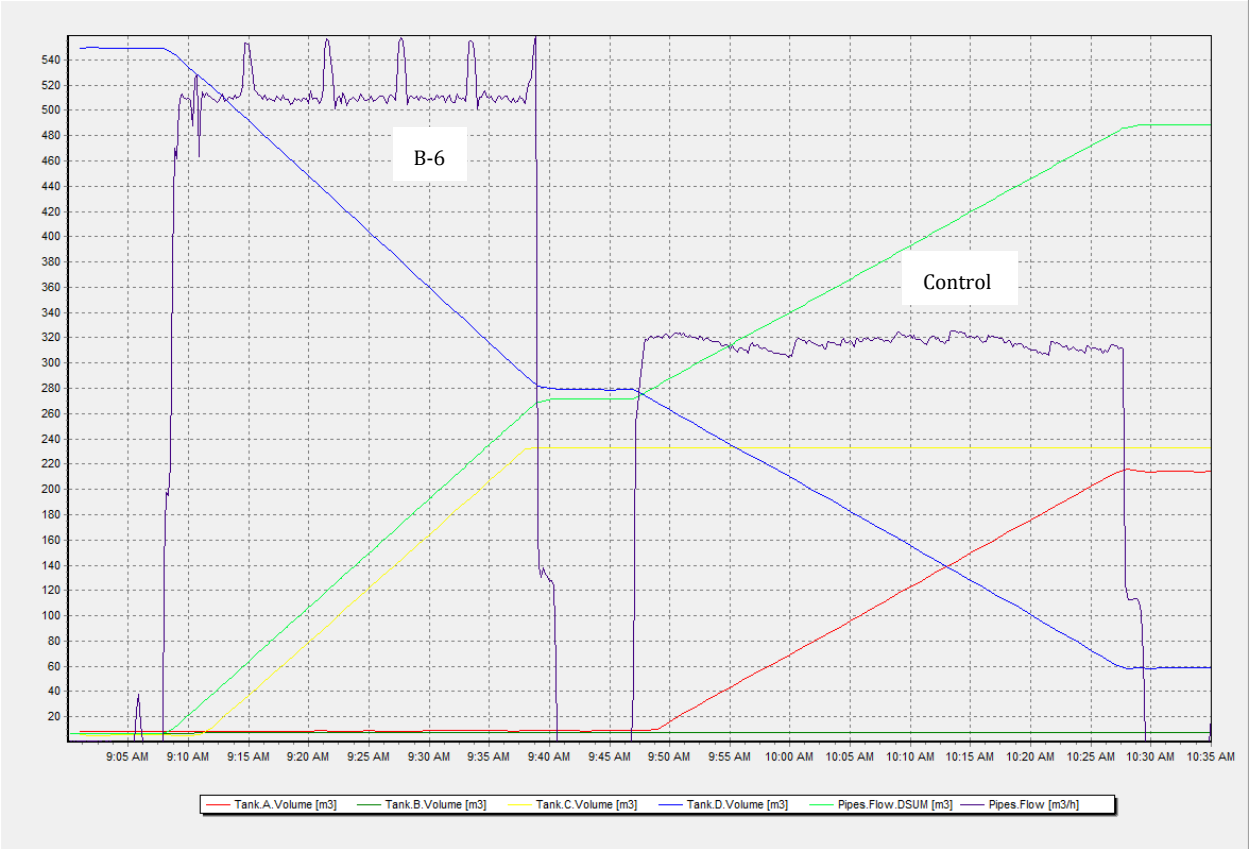


Figure B-9 BIO-SEA test cycle B-6 inlet

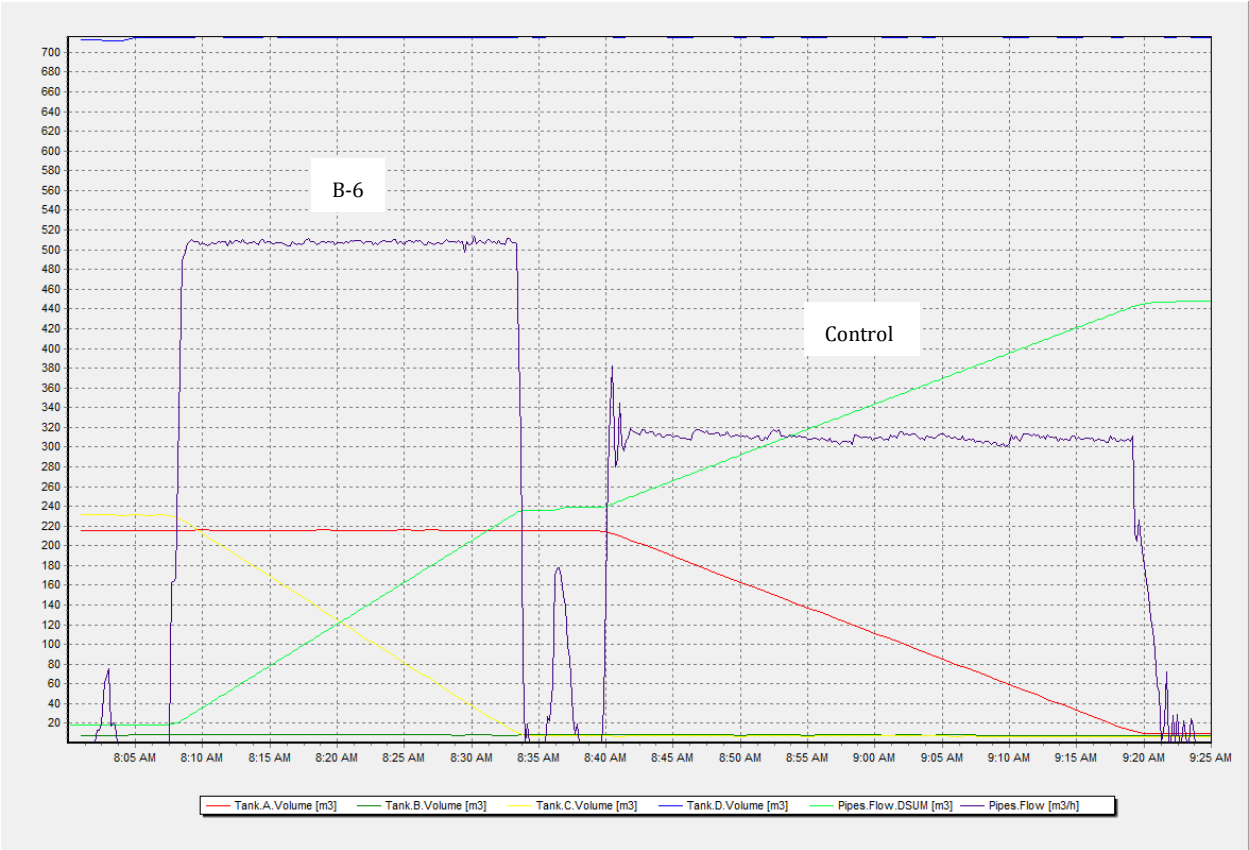


Figure B-10 BIO-SEA test cycle B-6 discharge

Table B-17 Test cycle M-3

Description	Data	Comments
Client treatment system	BIO-SEA	
Salinity (PSU)	33.9	
Retention tank no.	C	
Test cycle	M-3	
System cleaned before inlet	Yes	
Date and time inlet start	2011.10.20 09:04	
Date and time inlet stop	2011.10.20 09:31	
Flow during inlet (average)	507 m <sup>3</sup> /h	
Power consumption during inlet	78 kW	Provided by BIO-UV
Treated volume during inlet	224 m <sup>3</sup>	
App. volume used for treatment	236 m <sup>3</sup>	
System cleaned before discharge	Yes	
Date and time discharge start	2011.10.25 09:49	
Date and time discharge stop	2011.10.25 10:11	
Flow during discharge (average)	507 m <sup>3</sup> /h	
Power consumption during discharge	80 kW	Provided by BIO-UV
Treated volume during discharge	224 m <sup>3</sup>	
Use of cultured organisms	Yes	<i>Artemia and Tetraselmis</i>
Filter	Filtrex	
UV units	5	

Table B-18 Test cycle M-3

On-line measurements	DO sat. (%)	pH	Salinity	Temperature (C°)	Turbidity (NTU)
Control inlet (T0)	99.87±0.06	8.06±0.00	33.84±0.07	9.66±0.01	1.07±0.22
BIO-SEA inlet (T0)	100.15±0.32	8.04±0.01	33.86±0.00	9.71±0.01	1.70±0.60
BIO-SEA 1 <sup>st</sup> treatment (T0)	100.97±0.91	7.95±0.04	33.79±0.01	9.79±0.13	2.76±0.66
BIO-SEA inlet (T5)	89.55±0.94	7.89±0.00	33.85±0.01	9.66±0.00	1.28±1.50
BIO-SEA 2 <sup>nd</sup> treatment (T5)	91.76±1.44	7.79±0.02	33.78±0.01	9.77±0.01	1.44±0.45
Control discharge (T5)	89.66±0.36	7.86±0.01	33.85±0.02	9.95±0.00	1.75±1.29

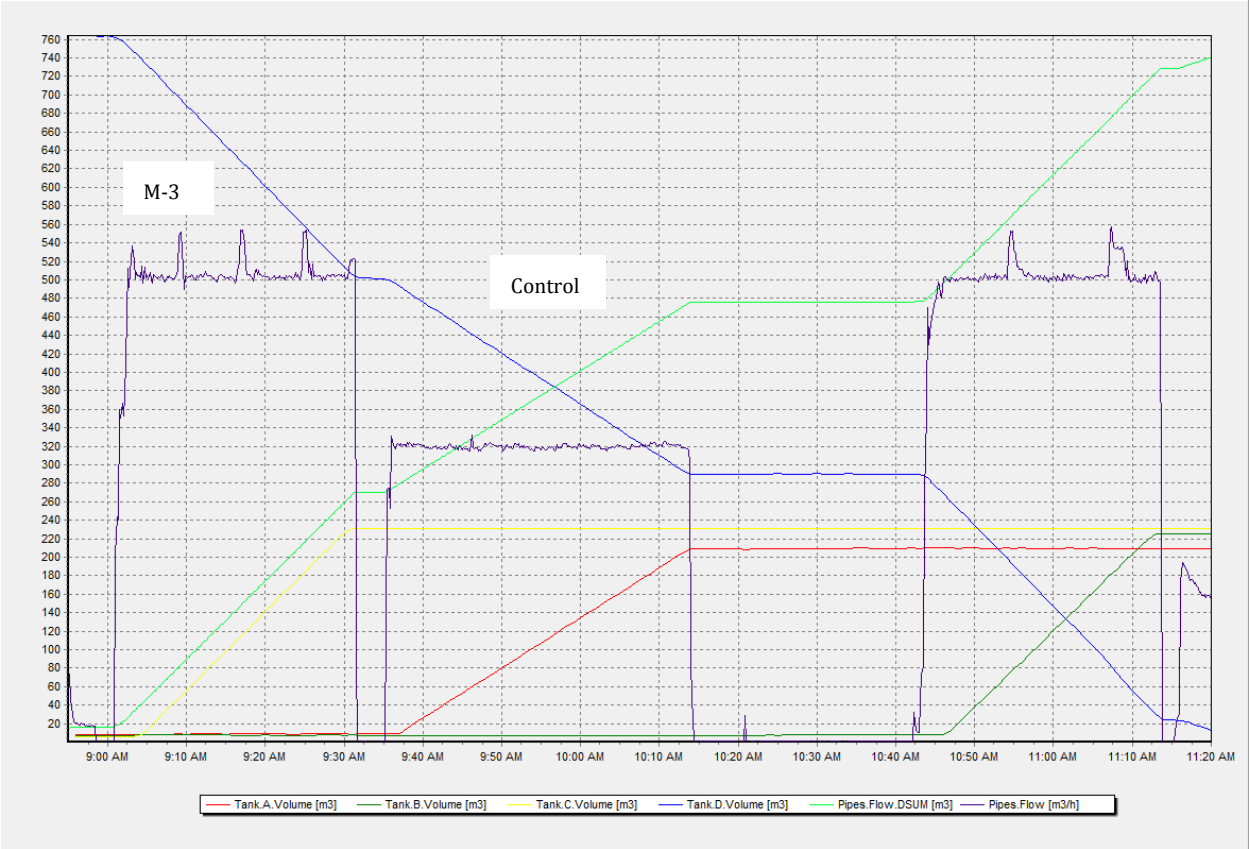


Figure B-11 BIO-SEA test cycles M-3 inlet

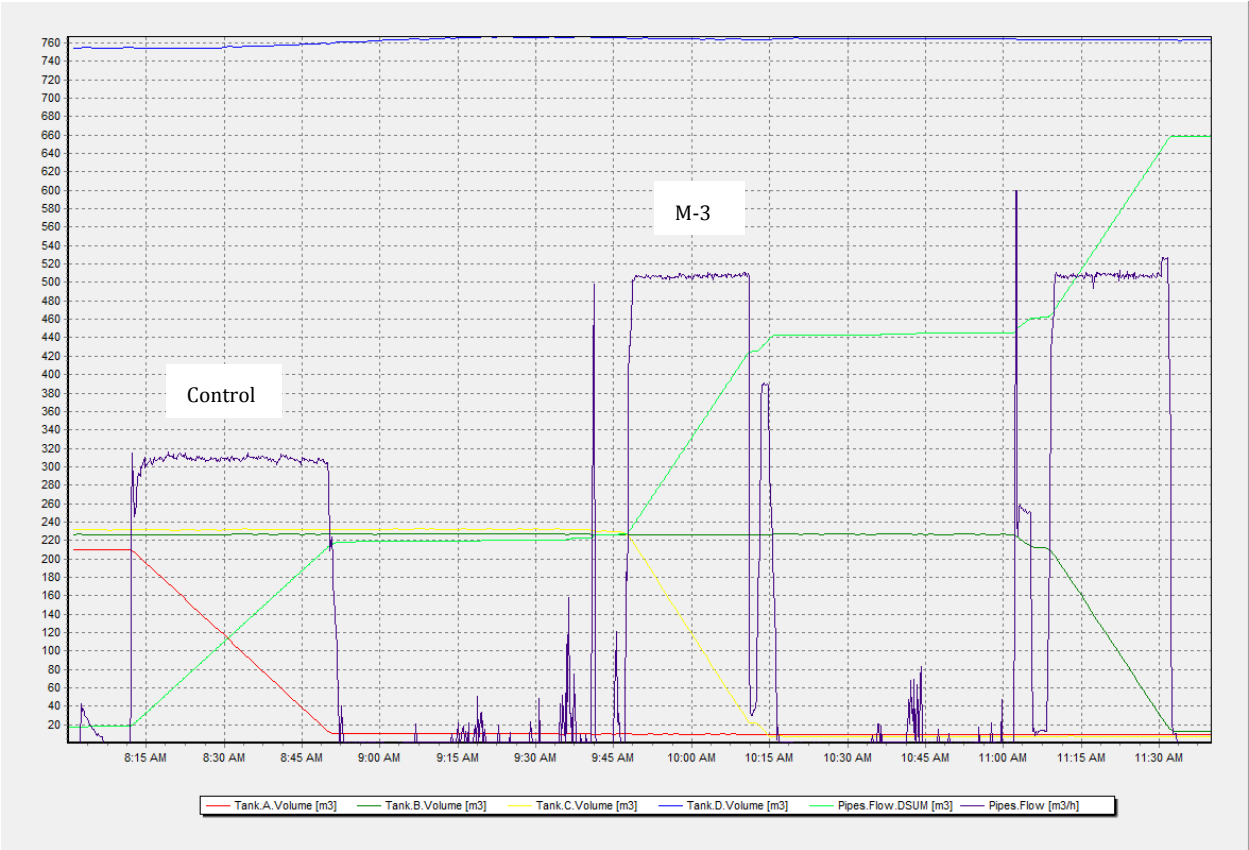


Figure B-12 BIO-SEA test cycles M-3 discharge

Table B-19 Test cycle M-4

Description	Data	Comments
Client treatment system	BIO-SEA	
Salinity (PSU)	36.4	
Retention tank no.	C	
Test cycle	M-4	
System cleaned before inlet	Yes	
Date and time inlet start	2011.10.27 09:09	
Date and time inlet stop	2011.10.27 09:35	
Flow during inlet (average)	508 m <sup>3</sup> /h	
Power consumption during inlet	80 kW	Provided by BIO-UV
Treated volume during inlet	220 m <sup>3</sup>	
App. volume used for treatment	228 m <sup>3</sup>	
System cleaned before discharge	Yes	
Date and time discharge start	2011.11.01 08:33	
Date and time discharge stop	2011.11.01 08:55	
Flow during discharge (average)	507 m <sup>3</sup> /h	
Power consumption during discharge	82 kW	Provided by BIO-UV
Treated volume during discharge	220 m <sup>3</sup>	
Use of cultured organisms	Yes	<i>Artemia and Tetraselmis</i>
Filter	Filtrex	
UV units	5	

Table B-20 Test cycle M-4

On-line measurements	DO sat. (%)	pH	Salinity	Temperature (C°)	Turbidity (NTU)
Control inlet (T0)	86.33±0.11	7.94±0.01	36.39±0.01	10.27±0.01	1.49±0.20
BIO-SEA inlet (T0)	86.34±0.18	7.93±0.01	36.40±0.00	10.32±0.00	2.22±0.67
BIO-SEA 1 <sup>st</sup> treatment (T0)	88.32±0.72	7.83±0.02	36.33±0.01	10.41±0.01	2.89±0.17
BIO-SEA inlet (T5)	84.73±0.80	7.81±0.00	36.36±0.01	10.38±0.01	1.02±1.04
BIO-SEA 2 <sup>nd</sup> treatment (T5)	84.63±0.54	7.75±0.01	36.29±0.01	10.50±0.00	1.50±0.24
Control discharge (T5)	80.71±2.00	7.83±0.01	36.28±0.03	10.33±0.00	0.62±2.14

Table B-21 Test cycle M-5

Description	Data	Comments
Client treatment system	BIO-SEA	
Salinity (PSU)	36.4	
Retention tank no.	B	
Test cycle	M-5	
System cleaned before inlet	Yes	
Date and time inlet start	2011.10.27 11:16	
Date and time inlet stop	2011.10.27 11:42	
Flow during inlet (average)	507 m <sup>3</sup> /h	
Power consumption during inlet	80 kW	Provided by BIO-UV
Treated volume during inlet	212 m <sup>3</sup>	
App. volume used for treatment	232 m <sup>3</sup>	
System cleaned before discharge	Yes	
Date and time discharge start	2011.11.01 10:28	
Date and time discharge stop	2011.11.01 10:50	
Flow during discharge (average)	507 m <sup>3</sup> /h	
Power consumption during discharge	82 kW	Provided by BIO-UV
Treated volume during discharge	212 m <sup>3</sup>	
Use of cultured organisms	Yes	<i>Artemia and Tetraselmis</i>
Filter	Filtrex	
UV units	5	

Table B-22 Test cycle M-5

On-line measurements	DO sat. (%)	pH	Salinity	Temperature (C°)	Turbidity (NTU)
Control inlet (T0)	86.33±0.11	7.94±0.01	36.39±0.01	10.27±0.01	1.49±0.20
BIO-SEA inlet (T0)	88.47±1.04	7.96±0.00	36.38±0.00	10.33±0.01	2.17±0.51
BIO-SEA 1 <sup>st</sup> treatment (T0)	89.99±0.95	7.88±0.00	36.32±0.01	10.44±0.01	2.93±0.07
BIO-SEA inlet (T5)	83.68±0.82	7.86±0.00	36.37±0.01	10.46±0.00	1.11±0.65
BIO-SEA 2 <sup>nd</sup> treatment (T5)	84.48±0.81	7.77±0.00	36.30±0.00	10.58±0.01	1.77±0.78
Control discharge (T5)	80.71±2.00	7.83±0.01	36.28±0.03	10.33±0.00	0.62±2.14

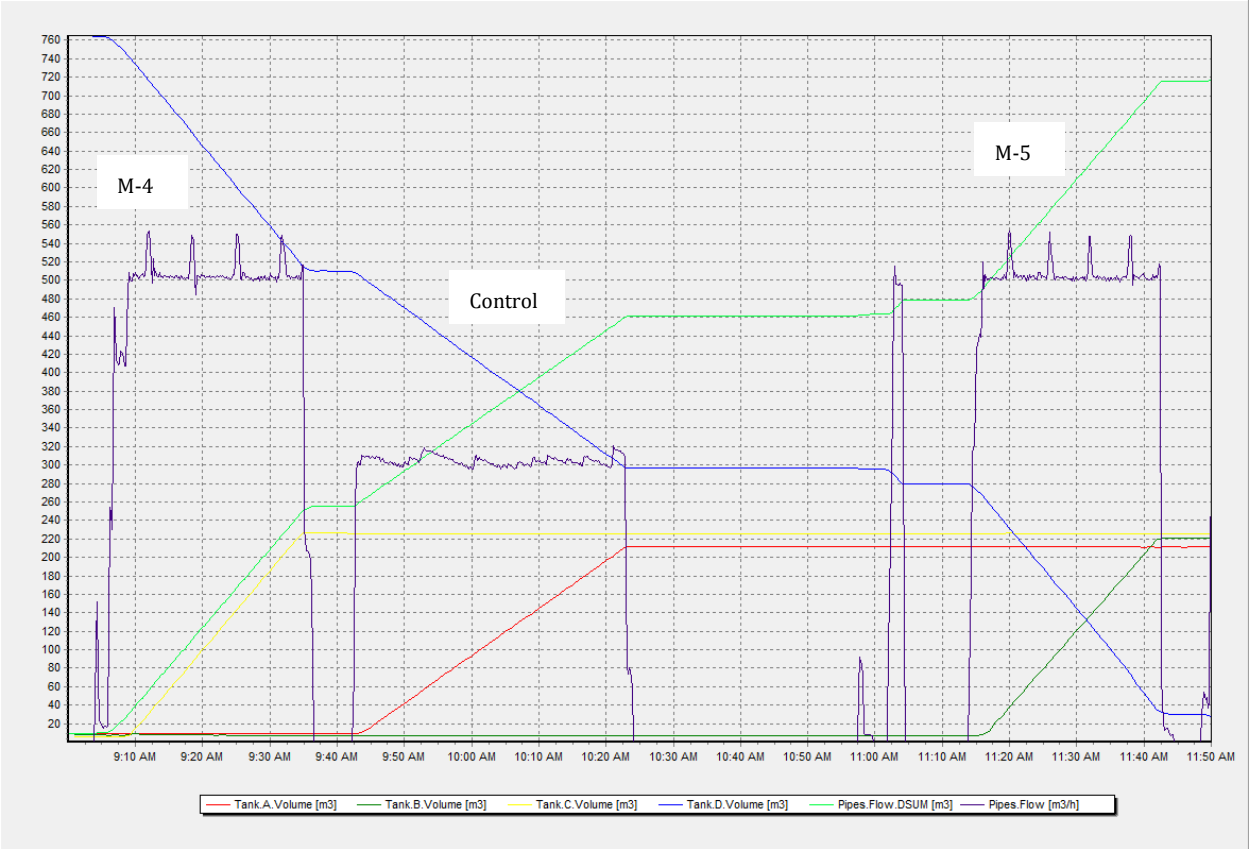


Figure B-13 BIO-SEA test cycles M-4 and M-5 inlet

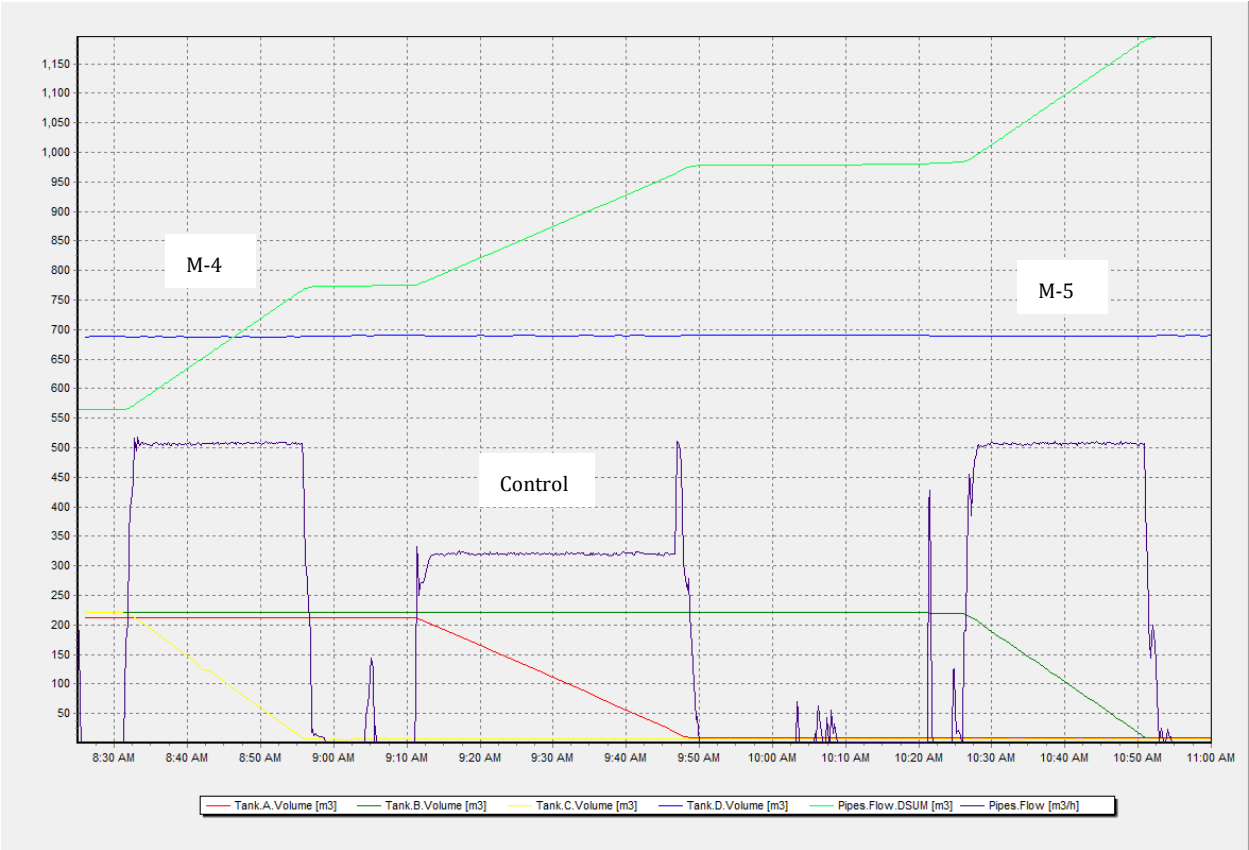


Figure B-14 BIO-SEA test cycles M-4 and M-5 discharge

## **A P P E N D I X   C**

Test results for physical and chemical properties of treated water and biological efficacy analysis in land-based testing

## Brackish water tests

Table C-1 Dissolved organic carbon (DOC) (mg/L)

Facility	Position	FR1	FR2	FR3	Average	SD	IMO
Test cycle B-1							
Control	Inlet	5.49	5.53	5.58	5.53	0.05	>5
BIO-SEA	Inlet	5.60	5.44	5.60	5.55	0.09	>5
BIO-SEA	2 <sup>nd</sup> treatment T5	5.42	5.29	5.18	5.30	0.12	-
Control	T5	5.04	5.03	4.90	4.99	0.08	-
Test cycle B-2							
Control	Inlet	5.49	5.53	5.58	5.53	0.05	>5
BIO-SEA	Inlet	5.54	5.57	5.51	5.54	0.03	>5
BIO-SEA	2 <sup>nd</sup> treatment T5	5.28	5.16	5.13	5.19	0.08	-
Control	T5	5.04	5.03	4.90	4.99	0.08	-
Test cycle B-3							
Control	Inlet	4.95	4.91	4.84	4.90	0.05	>5
BIO-SEA	Inlet	5.06	5.01	4.85	4.97	0.11	>5
BIO-SEA	2 <sup>nd</sup> treatment T5	5.06	5.01	4.98	5.02	0.04	-
Control	T5	4.45	4.57	4.44	4.48	0.07	-
Test cycle B-4							
Control	Inlet	4.95	4.91	4.84	4.90	0.05	>5
BIO-SEA	Inlet	4.88	4.88	4.89	4.88	0.005	>5
BIO-SEA	2 <sup>nd</sup> treatment T5	4.82	3.97	4.81	4.53	0.49	-
Control	T5	4.45	4.57	4.44	4.48	0.07	-
Test cycle B-5							
Control	Inlet	4.95	4.65	4.68	4.76	0.16	>5
BIO-SEA	Inlet	4.77	4.70	4.71	4.73	0.04	>5
BIO-SEA	2 <sup>nd</sup> treatment T5	4.14	4.26	4.32	4.24	0.09	-
Control	T5	4.28	4.17	4.22	4.22	0.05	-
Test cycle B-6							
Control	Inlet	6.11	6.11	5.95	6.06	0.09	>5
BIO-SEA	Inlet	5.91	6.21	5.73	5.95	0.24	>5
BIO-SEA	2 <sup>nd</sup> treatment T5	5.86	5.68	4.57	5.37	0.70	-
Control	T5	5.51	5.52	5.73	5.59	0.13	-

Table C-2 Particulate organic carbon (POC) (mg/L)

Facility	Position	FR1	FR2	FR3	Average	SD	IMO
Test cycle B-1							
Control	Inlet	6.35	6.11	6.09	6.18	0.15	>5
BIO-SEA	Inlet	6.25	6.24	6.29	6.26	0.03	>5
BIO-SEA	2 <sup>nd</sup> treatment T5	2.64	2.68	2.71	2.68	0.03	-
Control	T5	3.67	3.23	4.21	3.70	0.49	-
Test cycle B-2							
Control	Inlet	6.35	6.11	6.09	6.18	0.15	>5
BIO-SEA	Inlet	6.35	6.21	6.13	6.23	0.11	>5
BIO-SEA	2 <sup>nd</sup> treatment T5	2.58	2.75	2.84	2.73	0.13	-
Control	T5	3.67	3.23	4.21	3.70	0.49	-
Test cycle B-3							
Control	Inlet	6.31	6.57	6.22	6.37	0.18	>5
BIO-SEA	Inlet	6.04	6.04	5.93	6.00	0.06	>5
BIO-SEA	2 <sup>nd</sup> treatment T5	1.93	1.85	2.10	1.96	0.12	-
Control	T5	3.99	3.14	2.99	3.37	0.54	-
Test cycle B-4							
Control	Inlet	6.31	6.57	6.22	6.37	0.18	>5
BIO-SEA	Inlet	6.73	6.35	6.19	6.42	0.28	>5
BIO-SEA	2 <sup>nd</sup> treatment T5	2.36	3.55	2.47	2.79	0.66	-
Control	T5	3.99	3.14	2.99	3.37	0.54	-
Test cycle B-5							
Control	Inlet	6.34	6.74	6.38	6.49	0.22	>5
BIO-SEA	Inlet	5.63	6.26	6.50	6.13	0.45	>5
BIO-SEA	2 <sup>nd</sup> treatment T5	4.14	3.04	3.73	3.64	0.56	-
Control	T5	4.81	3.98	4.61	4.47	0.44	-
Test cycle B-6							
Control	Inlet	4.88	6.01	7.14	6.01	1.13	>5
BIO-SEA	Inlet	5.95	5.38	6.42	5.92	0.52	>5
BIO-SEA	2 <sup>nd</sup> treatment T5	2.45	2.23	3.60	2.76	0.74	-
Control	T5	3.02	2.73	2.68	2.81	0.19	-

Table C-3 Total suspended solids (TSS) (mg/L)

Facility	Position	FR1	FR2	FR3	Average	SD	IMO
Test cycle B-1							
Control	Inlet	54.7	55.7	53.0	54.5	1.38	>50
BIO-SEA	Inlet	54.7	55.3	54.1	54.7	0.61	>50
BIO-SEA	1 <sup>st</sup> treatment T0	44.8	53.4	51.0	49.7	4.43	-
BIO-SEA	2 <sup>nd</sup> treatment T5	30.5	28.5	27.4	28.8	1.56	-
Control	T5	41.2	36.0	36.4	37.9	2.93	-
Test cycle B-2							
Control	Inlet	54.7	55.7	53.0	54.5	1.38	>50
BIO-SEA	Inlet	49.2	55.1	61.2	55.2	5.99	>50
BIO-SEA	1 <sup>st</sup> treatment T0	48.8	54.7	55.8	53.1	3.78	-
BIO-SEA	2 <sup>nd</sup> treatment T5	31.6	32.4	31.2	31.7	0.59	-
Control	T5	41.2	36.0	36.4	37.9	2.93	-
Test cycle B-3							
Control	Inlet	47.5	48.2	51.2	49.0	1.95	>50
BIO-SEA	Inlet	51.1	50.8	49.0	50.3	1.15	>50
BIO-SEA	1 <sup>st</sup> treatment T0	48.9	49.0	50.1	49.3	0.66	-
BIO-SEA	2 <sup>nd</sup> treatment T5	26.6	27.4	26.0	26.7	0.67	-
Control	T5	43.3	41.1	34.5	39.6	4.56	-
Test cycle B-4							
Control	Inlet	47.5	48.2	51.2	49.0	1.95	>50
BIO-SEA	Inlet	51.4	49.9	48.8	50.1	1.32	>50
BIO-SEA	1 <sup>st</sup> treatment T0	49.5	48.6	50.0	49.4	0.69	-
BIO-SEA	2 <sup>nd</sup> treatment T5	30.1	29.2	28.2	29.1	0.96	-
Control	T5	43.3	41.1	34.5	39.6	4.56	-
Test cycle B-5							
Control	Inlet	-	60.8	63.8	62.3	2.12	>50
BIO-SEA	Inlet	55.8	56.6	58.5	57.0	1.37	>50
BIO-SEA	1 <sup>st</sup> treatment T0	55.0	57.7	57.2	56.6	1.46	-
BIO-SEA	2 <sup>nd</sup> treatment T5	46.1	40.1	38.5	41.6	4.01	-
Control	T5	48.1	45.7	-	46.9	1.70	-
Test cycle B-6							
Control	Inlet	59.1	55.7	59.0	57.9	1.94	>50
BIO-SEA	Inlet	55.9	55.2	55.2	55.4	0.44	>50
BIO-SEA	1 <sup>st</sup> treatment T0	54.7	53.7	57.0	55.1	1.71	-
BIO-SEA	2 <sup>nd</sup> treatment T5	45.1	40.2	40.6	42.0	2.72	-
Control	T5	47.8	45.8	42.8	45.5	2.47	-

Table C-4 Heterotrophic bacteria (CFU/mL)

Facility	Position	FR1	FR2	FR3	Average	SD	IMO
Test cycle B-1							
Control/ BIO-SEA	Inlet	54,100	31,800	64,500	50,133	16,707	>10,000
BIO-SEA	1 <sup>st</sup> treatment T0	99.0	50.0	36.5	61.8	32.9	-
BIO-SEA	2 <sup>nd</sup> treatment T5	160	79.0	49.0	95.8	57.1	-
Control	T5	108,750	108,000	127,000	114,583	10,760	-
Test cycle B-2							
Control/ BIO-SEA	Inlet	54,100	31,800	64,500	50,133	16,707	>10,000
BIO-SEA	1 <sup>st</sup> treatment T0	55.0	36.0	19.0	36.7	18.0	-
BIO-SEA	2 <sup>nd</sup> treatment T5	46.0	31.0	24.0	33.7	11.2	-
Control	T5	108,750	108,000	127,000	114,583	10,760	-
Test cycle B-3							
Control/ BIO-SEA	Inlet	26,150	33,600	40,900	33,550	7,315	>10,000
BIO-SEA	1 <sup>st</sup> treatment T0	216	130	140	162	47.1	-
BIO-SEA	2 <sup>nd</sup> treatment T5	255	145	295	232	77.7	-
Control	T5	227,000	245,000	236,500	236,167	9,005	-
Test cycle B-4							
Control/ BIO-SEA	Inlet	26,150	33,600	40,900	33,550	7,315	>10,000
BIO-SEA	1 <sup>st</sup> treatment T0	86.0	62.0	35.0	61.0	25.5	-
BIO-SEA	2 <sup>nd</sup> treatment T5	142	92.0	93.0	109	28.6	-
Control	T5	227,000	245,000	236,500	236,167	9,005	-
Test cycle B-5							
Control/ BIO-SEA	Inlet	47,950	48,600	42,700	46,417	3235	>10,000
BIO-SEA	1 <sup>st</sup> treatment T0	22.0	20.0	19.0	20.3	1.53	-
BIO-SEA	2 <sup>nd</sup> treatment T5	45.0	28.0	24.0	32.3	11.2	-
Control	T5	114,000	118,000	128,000	120,000	7211	-
Test cycle B-6							
Control/ BIO-SEA	Inlet	37,700	44,100	43,200	41,667	3,465	>10,000
BIO-SEA	1 <sup>st</sup> treatment T0	92.0	123	85.0	99.8	19.9	-
BIO-SEA	2 <sup>nd</sup> treatment T5	112	70.0	43.0	75.0	34.8	-
Control	T5	>200,000	>200,000	>200,000	>200,000	0	-

Table C-5 *E. coli* (CFU/100 mL)

Facility	Position	FR1	FR2	FR3	Average	SD	IMO
Test cycle B-1							
Control/ BIO-SEA	Inlet	10.00	4.50	8.00	7.50	2.78	-
BIO-SEA	2 <sup>nd</sup> treatment T5	<1.00	<1.00	<1.00	<1.00	0.00	<250
Control	T5	12.00	12.00	26.00	16.67	8.08	-
Test cycle B-2							
Control/ BIO-SEA	Inlet	10.00	4.50	8.00	7.50	2.78	-
BIO-SEA	2 <sup>nd</sup> treatment T5	<1.00	<1.00	<1.00	<1.00	0.00	<250
Control	T5	12.00	12.00	26.00	16.67	8.08	-
Test cycle B-3							
Control/ BIO-SEA	Inlet	12.00	6.50	18.0	12.17	5.75	-
BIO-SEA	2 <sup>nd</sup> treatment T5	<1.00	<1.00	<1.00	<1.00	0.00	<250
Control	T5	34.00	18.00	24.00	25.33	8.08	-
Test cycle B-4							
Control/ BIO-SEA	Inlet	12.00	6.50	18.0	12.17	5.75	-
BIO-SEA	2 <sup>nd</sup> treatment T5	<1.00	<1.00	<1.00	<1.00	0.00	<250
Control	T5	34.00	18.00	24.00	25.33	8.08	-
Test cycle B-5							
Control/ BIO-SEA	Inlet	34.00	44.5	28.0	35.50	8.35	-
BIO-SEA	2 <sup>nd</sup> treatment T5	<1.00	<1.00	<1.00	<1.00	0.00	<250
Control	T5	93.00	70.00	77.00	80.00	11.79	-
Test cycle B-6							
Control/ BIO-SEA	Inlet	212.00	210.00	308.00	243.33	56.01	-
BIO-SEA	2 <sup>nd</sup> treatment T5	<1.00	<1.00	<1.00	<1.00	0.00	<250
Control	T5	2,420	2,420	2,420	2,420	0	-

Table C-6 Enterococci (CFU/100 mL)

Facility	Position	FR1	FR2	FR3	Average	SD	IMO
Test cycle B-1							
Control/ BIO-SEA	Inlet	>2,420	>2,420	1,046	1,962	793.28	-
BIO-SEA	2 <sup>nd</sup> treatment T5	2.00	<1.00	<1.00	1.33	0.58	<100
Control	T5	727	1,553	596	959	519	-
Test cycle B-2							
Control/ BIO-SEA	Inlet	>2,420	>2,420	1,046	1,962	793.28	-
BIO-SEA	2 <sup>nd</sup> treatment T5	<1.00	<1.00	<1.00	<1.00	0.00	<100
Control	T5	727	1,553	596	959	519	-
Test cycle B-3							
Control/ BIO-SEA	Inlet	461	579	435	491.67	76.74	-
BIO-SEA	2 <sup>nd</sup> treatment T5	<1.00	<1.00	<1.00	<1.00	0.00	<100
Control	T5	1,083	1,203	980	1,089	112	-
Test cycle B-4							
Control/ BIO-SEA	Inlet	461	579	435	491.67	76.74	-
BIO-SEA	2 <sup>nd</sup> treatment T5	<1.00	<1.00	<1.00	<1.00	0.00	<100
Control	T5	1,083	1,203	980	1,089	112	-
Test cycle B-5							
Control/ BIO-SEA	Inlet	90	345	579	338	244.58	-
BIO-SEA	2 <sup>nd</sup> treatment T5	<1.00	<1.00	<1.00	<1.00	0.00	<100
Control	T5	58	42	35	45	12	-
Test cycle B-6							
Control/ BIO-SEA	Inlet	130	579	275	328	229.14	
BIO-SEA	2 <sup>nd</sup> treatment T5	<1.00	<1.00	<1.00	<1.00	0.00	<100
Control	T5	61	56	58	58	2	

Table C-7 *Vibrio cholerae* (CFU/100 mL)

Facility	Position	FR1	FR2	FR3	Average	SD	IMO
Test cycle B-1							
BIO-SEA	2 <sup>nd</sup> treatment T5	Negative	Negative	Negative	Negative	-	<1
Control	T5	Negative	Negative	Negative	Negative	-	-
Test cycle B-2							
BIO-SEA	2 <sup>nd</sup> treatment T5	Negative	Negative	Negative	Negative	-	<1
Control	T5	Negative	Negative	Negative	Negative	-	-
Test cycle B-3							
BIO-SEA	2 <sup>nd</sup> treatment T5	Negative	Negative	Negative	Negative	-	<1
Control	T5	Negative	Negative	Negative	Negative	-	-
Test cycle B-4							
BIO-SEA	2 <sup>nd</sup> treatment T5	Negative	Negative	Negative	Negative	-	<1
Control	T5	Negative	Negative	Negative	Negative	-	-
Test cycle B-5							
BIO-SEA	2 <sup>nd</sup> treatment T5	Negative	Negative	Negative	Negative	-	<1
Control	T5	Negative	Negative	Negative	Negative	-	-
Test cycle B-6							
BIO-SEA	2 <sup>nd</sup> treatment T5	Negative	Negative	Negative	Negative	-	<1
Control	T5	Negative	Negative	Negative	Negative	-	-

Table C-8 Primary production (DPM)

Facility	Position	FR1	FR2	FR3	Average	SD
Test cycle B-1						
Control/ BIO-SEA	Inlet	5,475	4,585	4,879	4,979.3	453.9
BIO-SEA	1 <sup>st</sup> treatment T0	24.1	20.7	24.6	23.1	2.1
BIO-SEA	2 <sup>nd</sup> treatment T5	2.19	0.25	0.00E+00	0.81	1.2
Control	T5	577	639	839	685	137
Test cycle B-2						
Control/ BIO-SEA	Inlet	5,475	4,585	4,879	4,979.3	453.9
BIO-SEA	1 <sup>st</sup> treatment T0	42.1	45.7	54.3	47.4	6.31
BIO-SEA	2 <sup>nd</sup> treatment T5	2.85	0.44	0.00E+00	1.09	1.53
Control	T5	577	639	839	685	137
Test cycle B-3						
Control/ BIO-SEA	Inlet	5,424	4,847	5,370	5,213.7	318.7
BIO-SEA	1 <sup>st</sup> treatment T0	75.5	60.7	62.3	66.1	8.13
BIO-SEA	2 <sup>nd</sup> treatment T5	0.28	0.00	0.00	0.09	0.16
Control	T5	897	858	1,348	1,034	272
Test cycle B-4						
Control/ BIO-SEA	Inlet	5,424	4,847	5,370	5,213.7	318.7
BIO-SEA	1 <sup>st</sup> treatment T0	187	147	150	161	22.1
BIO-SEA	2 <sup>nd</sup> treatment T5	0.81	0.00	0.97	0.59	0.52
Control	T5	897	858	1,348	1,034	272
Test cycle B-5						
Control/ BIO-SEA	Inlet	3,507	3,342	3,422	3,423.7	82.5
BIO-SEA	1 <sup>st</sup> treatment T0	98	66.8	77	80.6	15.9
BIO-SEA	2 <sup>nd</sup> treatment T5	7.56	5.22	19.5	10.8	7.67
Control	T5	1,208	1,265	1,216	1,230	31.3
Test cycle B-6						
Control/ BIO-SEA	Inlet	1,691	1,459	1,584	1,578	116.1
BIO-SEA	1 <sup>st</sup> treatment T0	42.8	47.4	42.9	44.4	2.67
BIO-SEA	2 <sup>nd</sup> treatment T5	1.55	2.18	1.82	1.85	0.32
Control	T5	1,301	1,056	1,068	1,142	138

Table C-9 Number of organisms  $\geq 10$  and  $< 50 \mu\text{m}$  (Lugol's solution) (organisms/mL)

Facility	Position	FR1	FR2	FR3	Average	SD	IMO
Test cycle B-1							
Control/ BIO-SEA	Inlet	908	1,520	921	1,116	349.6	>1,000
Test cycle B-2							
Control/ BIO-SEA	Inlet	908	1,520	921	1,116	349.6	>1,000
Test cycle B-3							
Control/ BIO-SEA	Inlet	5,140	4,202	4,649	4,664	469.2	>1,000
Test cycle B-4							
Control/ BIO-SEA	Inlet	5,140	4,202	4,649	4,664	469.2	>1,000
Test cycle B-5							
Control/ BIO-SEA	Inlet	1,895	1,910	1,937	1,914	21.3	>1,000
Test cycle B-6							
Control/ BIO-SEA	Inlet	1,147	995	984	1,042	91.1	>1,000

Table C-10 Number of organisms  $\geq 10$  and  $< 50 \mu\text{m}$  (MPN) in brackish water (organisms/mL)

Facility	Position	FR1	FR2	FR3	Average	SD	IMO
Test cycle B-1							
Control/ BIO-SEA	Inlet	1,300	9,200	16,000	8,833	7,356.9	$>1,000$
BIO-SEA	2 <sup>nd</sup> treatment T5	$<0.18$	$<0.18$	$<0.18$	$<0.18$	0.00	$<10$
Control	T5	240	170	280	230	55.68	$>100$
Test cycle B-2							
Control/ BIO-SEA	Inlet	1,300	9,200	16,000	8,833	7,356.9	$>1,000$
BIO-SEA	2 <sup>nd</sup> treatment T5	$<0.18$	$<0.18$	$<0.18$	$<0.18$	0.00	$<10$
Control	T5	240	170	280	230	55.68	$>100$
Test cycle B-3							
Control/ BIO-SEA	Inlet	3,500	3,500	5,400	4,133	1,097	$>1000$
BIO-SEA	2 <sup>nd</sup> treatment T5	$<0.18$	$<0.18$	$<0.18$	$<0.18$	0.00	$<10$
Control	T5	1,600	$>1,600$	$>1,600$	$>1,600$	0.00	$>100$
Test cycle B-4							
Control/ BIO-SEA	Inlet	3,500	3,500	5,400	4,133	1097	$>1,000$
BIO-SEA	2 <sup>nd</sup> treatment T5	0.20	$<0.18$	$<0.18$	0.19	0.01	$<10$
Control	T5	1,600	$>1,600$	$>1,600$	$>1,600$	0.00	$>100$
Test cycle B-5							
Control/ BIO-SEA	Inlet	9,200	5,400	9,200	7,933	2,193.9	$>1,000$
BIO-SEA	2 <sup>nd</sup> treatment T5	0.20	$<0.18$	$<0.18$	0.19	0.01	$<10$
Control	T5	$>1,600$	1,600	540	1,247	611.99	$>100$
Test cycle B-6							
Control/ BIO-SEA	Inlet	$>16,000$	2,400	9,200	9,200	6,800	$>1,000$
BIO-SEA	2 <sup>nd</sup> treatment T5	0.68	0.40	0.45	0.51	0.15	$<10$
Control	T5	$>1,600$	1,600	$>1,600$	1,600	0.00	$>100$

Table C-11 Number of organisms  $\geq 50 \mu\text{m}$  (organisms/ $\text{m}^3$ )

Facility	Position	FR1	FR2	FR3	Average	SD	IMO
Test cycle B-1							
Control	Inlet	126,667	187,067	86,167	133,300	50,776	>100,000
BIO-SEA	Inlet	77,333	185,500	218,500	160,445	73,843	>100,000
BIO-SEA	1 <sup>st</sup> treatment T0	36.0	32.0	25.0	31.0	5.57	-
BIO-SEA	2 <sup>nd</sup> treatment T5	0.00	0.00	1.25	0.42	0.72	<10
Control	T5	48,960	53,250	51,100	51,103	2,145	>100
Test cycle B-2							
Control	Inlet	126,667	187,067	86,167	133,300	50,776	>100,000
BIO-SEA	Inlet	200,667	73,500	129,000	134,389	63,754	>100,000
BIO-SEA	1 <sup>st</sup> treatment T0	31.4	55.0	65.0	50.5	17.2	-
BIO-SEA	2 <sup>nd</sup> treatment T5	1.25	0.00	0.00	0.42	0.72	<10
Control	T5	48,960	53,250	51,100	51,103	2,145	>100
Test cycle B-3							
Control	Inlet	156,100	139,747	121,018	138,955	17,554	>100,000
BIO-SEA	Inlet	159,467	152,000	149,120	153,529	5,340	>100,000
BIO-SEA	1 <sup>st</sup> treatment T0	40.0	43.3	63.3	48.9	12.6	-
BIO-SEA	2 <sup>nd</sup> treatment T5	3.33	3.33	3.33	3.33	0.00	<10
Control	T5	45,240	35,933	25,333	35,502	9,960	>100
Test cycle B-4							
Control	Inlet	156,100	139,747	121,018	138,955	17,554	>100,000
BIO-SEA	Inlet	112,618	178,400	153,520	148,179	33,215	>100,000
BIO-SEA	1 <sup>st</sup> treatment T0	26.7	30.0	33.3	30.0	3.33	-
BIO-SEA	2 <sup>nd</sup> treatment T5	3.33	0.00	6.67	3.33	3.33	<10
Control	T5	45,240	35,933	25,333	35,502	9,960	>100
Test cycle B-5							
Control	Inlet	131,487	131,580	275,000	179,356	82,831	>100,000
BIO-SEA	Inlet	203,840	138,960	179,920	174,240	32,811	>100,000
BIO-SEA	1 <sup>st</sup> treatment T0	35.0	45.0	15.0	31.7	15.3	-
BIO-SEA	2 <sup>nd</sup> treatment T5	0.00	0.00	0.00	0.00	0.00	<10
Control	T5	71,458	80,654	68,024	73,379	6,530	>100
Test cycle B-6							
Control	Inlet	140,533	135,700	151,300	142,511	7,986	>100,000
BIO-SEA	Inlet	163,773	146,160	149,100	153,011	9,436	>100,000
BIO-SEA	1 <sup>st</sup> treatment T0	45.0	30.0	22.5	32.5	11.5	-
BIO-SEA	2 <sup>nd</sup> treatment T5	0.00	0.00	5.00	1.67	2.89	<10
Control	T5	92,033	78,400	82,800	84,411	6,958	>100

## Marine water tests

Table C-12 Dissolved organic carbon (DOC) (mg/L)

Facility	Position	FR1	FR2	FR3	Average	SD	IMO
Test cycle M-1							
Control	Inlet	3.35	3.24	3.22	3.27	0.07	>1
BIO-SEA	Inlet	3.32	3.22	3.15	3.23	0.09	>1
BIO-SEA	2 <sup>nd</sup> treatment T5	3.55	3.57	3.56	3.56	0.008	-
Control	T5	3.45	3.42	3.42	3.43	0.02	-
Test cycle M-2							
Control	Inlet	3.35	3.24	3.22	3.27	0.07	>1
BIO-SEA	Inlet	3.22	3.20	3.28	3.23	0.04	>1
BIO-SEA	2 <sup>nd</sup> treatment T5	3.41	3.68	3.63	3.57	0.15	-
Control	T5	3.45	3.42	3.42	3.43	0.02	-
Test cycle M-3							
Control	Inlet	3.21	3.37	2.39	2.99	0.53	>1
BIO-SEA	Inlet	3.39	3.44	2.62	3.15	0.46	>1
BIO-SEA	2 <sup>nd</sup> treatment T5	2.49	2.53	2.62	2.54	0.07	-
Control	T5	2.32	3.30	3.44	3.02	0.61	-
Test cycle M-4							
Control	Inlet	2.41	1.94	2.05	2.13	0.25	>1
BIO-SEA	Inlet	2.46	2.18	2.20	2.28	0.16	>1
BIO-SEA	2 <sup>nd</sup> treatment T5	2.51	2.22	2.41	2.38	0.15	-
Control	T5	2.48	2.24	2.40	2.37	0.12	-
Test cycle M-5							
Control	Inlet	2.41	1.94	2.05	2.13	0.25	>1
BIO-SEA	Inlet	2.08	2.20	2.15	2.14	0.06	>1
BIO-SEA	2 <sup>nd</sup> treatment T5	2.40	2.31	2.40	2.37	0.05	-
Control	T5	2.48	2.24	2.40	2.37	0.12	-

Table C-13 Particulate organic carbon (POC) (mg/L)

Facility	Position	FR1	FR2	FR3	Average	SD	IMO
Test cycle M-1							
Control	Inlet	2.59	2.82	2.75	2.72	0.12	>1
BIO-SEA	Inlet	2.57	2.82	3.05	2.82	0.24	>1
BIO-SEA	2 <sup>nd</sup> treatment T5	0.42	0.41	0.37	0.40	0.03	-
Control	T5	0.47	0.67	0.49	0.55	0.11	-
Test cycle M-2							
Control	Inlet	2.59	2.82	2.75	2.72	0.12	>1
BIO-SEA	Inlet	2.66	2.93	2.92	2.84	0.16	>1
BIO-SEA	2 <sup>nd</sup> treatment T5	0.67	0.32	0.41	0.47	0.18	-
Control	T5	0.47	0.67	0.49	0.55	0.11	-
Test cycle M-3							
Control	Inlet	3.18	2.84	3.80	3.27	0.49	>1
BIO-SEA	Inlet	2.70	2.93	3.63	3.09	0.49	>1
BIO-SEA	2 <sup>nd</sup> treatment T5	2.10	1.95	1.82	1.96	0.14	-
Control	T5	2.52	1.43	1.55	1.84	0.60	-
Test cycle M-4							
Control	Inlet	2.65	3.27	2.89	2.94	0.31	>1
BIO-SEA	Inlet	2.72	2.77	2.83	2.77	0.06	>1
BIO-SEA	2 <sup>nd</sup> treatment T5	0.65	0.87	0.75	0.76	0.11	-
Control	T5	0.76	0.91	0.99	0.89	0.12	-
Test cycle M-5							
Control	Inlet	2.65	3.27	2.89	2.94	0.31	>1
BIO-SEA	Inlet	3.08	2.81	2.86	2.91	0.14	>1
BIO-SEA	2 <sup>nd</sup> treatment T5	0.95	1.06	1.06	1.02	0.07	-
Control	T5	0.76	0.91	0.99	0.89	0.12	-

Table C-14 Total suspended solids (TSS) (mg/L)

Facility	Position	FR1	FR2	FR3	Average	SD	IMO
Test cycle M-1							
Control	Inlet	14.5	13.9	11.7	13.4	1.51	>1
BIO-SEA	Inlet	12.1	12.6	12.2	12.3	0.26	>1
BIO-SEA	1 <sup>st</sup> treatment T0	12.0	12.6	12.9	12.5	0.47	-
BIO-SEA	2 <sup>nd</sup> treatment T5	4.77	4.90	4.67	4.78	0.11	-
Control	T5	4.84	3.76	3.71	4.1	0.64	-
Test cycle M-2							
Control	Inlet	14.5	13.9	11.7	13.4	1.51	>1
BIO-SEA	Inlet	14.5	12.3	13.6	13.5	1.13	>1
BIO-SEA	1 <sup>st</sup> treatment T0	12.2	13.6	14.1	13.3	0.96	-
BIO-SEA	2 <sup>nd</sup> treatment T5	5.36	6.11	5.60	5.69	0.38	-
Control	T5	4.84	3.76	3.71	4.1	0.64	-
Test cycle M-3							
Control	Inlet	9.6	9.8	8.79	9.39	0.53	>1
BIO-SEA	Inlet	8.51	9.59	8.83	8.98	0.55	>1
BIO-SEA	1 <sup>st</sup> treatment T0	8.45	8.21	6.94	7.87	0.81	-
BIO-SEA	2 <sup>nd</sup> treatment T5	4.02	4.62	2.93	3.86	0.86	-
Control	T5	3.67	3.54	4.62	3.94	0.59	-
Test cycle M-4							
Control	Inlet	9.44	10.0	7.54	8.99	1.29	>1
BIO-SEA	Inlet	9.49	10.2	9.70	9.80	0.37	>1
BIO-SEA	1 <sup>st</sup> treatment T0	10.2	8.59	8.60	9.13	0.93	-
BIO-SEA	2 <sup>nd</sup> treatment T5	3.2	3.4	3.73	3.44	0.27	-
Control	T5	4.27	3.6	6	4.62	1.24	-
Test cycle M-5							
Control	Inlet	9.44	10.0	7.54	8.99	1.29	>1
BIO-SEA	Inlet	10.4	10.9	9.39	10.2	0.77	>1
BIO-SEA	1 <sup>st</sup> treatment T0	9.25	8.50	10.3	9.35	0.91	-
BIO-SEA	2 <sup>nd</sup> treatment T5	4.67	3.4	3.2	3.76	0.8	-
Control	T5	4.27	3.6	6	4.62	1.24	-

Table C-15 Heterotrophic bacteria (CFU/mL)

Facility	Position	FR1	FR2	FR3	Average	SD	IMO
Test cycle M-1							
Control/ BIO-SEA	Inlet	223,000	295,000	265,000	261,000	36,166.3	>10,000
BIO-SEA	1 <sup>st</sup> treatment T0	205	69.0	54.0	109	83.2	-
BIO-SEA	2 <sup>nd</sup> treatment T5	90.0	54.0	26.0	56.7	32.1	-
Control	T5	186,000	194,000	210,000	196,667	12,220	-
Test cycle M-2							
Control/ BIO-SEA	Inlet	223,000	295,000	265,000	261,000	36,166.3	>10,000
BIO-SEA	1 <sup>st</sup> treatment T0	71.0	43.0	35.0	49.7	18.9	-
BIO-SEA	2 <sup>nd</sup> treatment T5	56.0	48.0	25.0	43.0	16.1	-
Control	T5	186,000	194,000	210,000	196,667	12,220	-
Test cycle M-3							
Control/ BIO-SEA	Inlet	755,000	722,500	825,000	767,500	52,380.8	>10,000
BIO-SEA	1 <sup>st</sup> treatment T0	733	121	230	361	326	-
BIO-SEA	2 <sup>nd</sup> treatment T5	39.0	21.0	20.0	26.7	10.7	-
Control	T5	122,750	99,500	139,000	120,417	19,853	-
Test cycle M-4							
Control/ BIO-SEA	Inlet	114,000	124,500	131,000	123,167	8,578.1	>10,000
BIO-SEA	1 <sup>st</sup> treatment T0	178	100	64.0	114	58.3	-
BIO-SEA	2 <sup>nd</sup> treatment T5	94.0	20.0	18.0	44.0	43.3	-
Control	T5	139,000	145,500	160,000	148,167	10,751	-
Test cycle M-5							
Control/ BIO-SEA	Inlet	114,000	124,500	131,000	123,167	8,578.1	>10,000
BIO-SEA	1 <sup>st</sup> treatment T0	35.0	35.0	28.0	32.7	4.04	-
BIO-SEA	2 <sup>nd</sup> treatment T5	25.0	13.0	18.0	18.7	6.03	-
Control	T5	139,000	145,500	160,000	148,167	10,751	-

Table C-16 *E. coli* (CFU/100 mL)

Facility	Position	FR1	FR2	FR3	Average	SD	IMO
Test cycle M-1							
Control/ BIO-SEA	Inlet	138.00	87.5	137	120.83	28.87	-
BIO-SEA	2 <sup>nd</sup> treatment T5	<1.00	<1.00	<1.00	<1.00	0.00	<250
Control	T5	36.50	50.00	25.00	37.17	12.51	-
Test cycle M-2							
Control/ BIO-SEA	Inlet	138.00	87.5	137	120.83	28.87	-
BIO-SEA	2 <sup>nd</sup> treatment T5	<1.00	<1.00	<1.00	<1.00	0.00	<250
Control	T5	36.50	50.00	25.00	37.17	12.51	-
Test cycle M-3							
Control/ BIO-SEA	Inlet	816.00	631	579	675.33	124.56	-
BIO-SEA	2 <sup>nd</sup> treatment T5	<1.00	<1.00	<1.00	<1.00	0.00	<250
Control	T5	67.50	83.00	56.00	68.83	13.55	-
Test cycle M-4							
Control/ BIO-SEA	Inlet	185.00	172	191	182.67	9.71	-
BIO-SEA	2 <sup>nd</sup> treatment T5	<1.00	<1.00	<1.00	<1.00	0.00	<250
Control	T5	36.00	50.50	46.00	44.17	7.42	-
Test cycle M-5							
Control/ BIO-SEA	Inlet	185.00	172	191	182.67	9.71	-
BIO-SEA	2 <sup>nd</sup> treatment T5	<1.00	<1.00	<1.00	<1.00	0.00	<250
Control	T5	36.00	50.50	46.00	44.17	7.42	-

Table C-17 Enterococci (CFU/100 mL)

Facility	Position	FR1	FR2	FR3	Average	SD	IMO
Test cycle M-1							
Control/ BIO-SEA	Inlet	248	517	457	407	141	-
BIO-SEA	2 <sup>nd</sup> treatment T5	<1.00	<1.00	<1.00	<1.00	0.00	<100
Control	T5	179	152	99	143	41	-
Test cycle M-2							
Control/ BIO-SEA	Inlet	248	517	457	407	141	-
BIO-SEA	2 <sup>nd</sup> treatment T5	<1.00	<1.00	<1.00	<1.00	0.00	<100
Control	T5	179	152	99	143	41	-
Test cycle M-3							
Control/ BIO-SEA	Inlet	1,986	1,733	1,309	1,676	342	-
BIO-SEA	2 <sup>nd</sup> treatment T5	<1.00	<1.00	<1.00	<1.00	0.00	<100
Control	T5	>2,420	>2,420	1,553	2,131	501	-
Test cycle M-4							
Control/ BIO-SEA	Inlet	387	1,092	687	722	354	-
BIO-SEA	2 <sup>nd</sup> treatment T5	<1.00	<1.00	<1.00	<1.00	0.00	<100
Control	T5	205	210	172	196	21	-
Test cycle M-5							
Control/ BIO-SEA	Inlet	387	1,092	687	722	354	-
BIO-SEA	2 <sup>nd</sup> treatment T5	<1.00	<1.00	<1.00	<1.00	0.00	<100
Control	T5	205	210	172	196	21	-

Table C-18 *Vibrio cholerae* (CFU/100 mL)

Facility	Position	FR1	FR2	FR3	Average	SD	IMO
Test cycle M-1							
BIO-SEA	2 <sup>nd</sup> treatment T5	Negative	Negative	Negative	Negative	-	<1
Control	T5	Negative	Negative	Negative	Negative	-	-
Test cycle M-2							
BIO-SEA	2 <sup>nd</sup> treatment T5	Negative	Negative	Negative	Negative	-	<1
Control	T5	Negative	Negative	Negative	Negative	-	-
Test cycle M-3							
BIO-SEA	2 <sup>nd</sup> treatment T5	Negative	Negative	Negative	Negative	-	<1
Control	T5	Negative	Negative	Negative	Negative	-	-
Test cycle M-4							
BIO-SEA	2 <sup>nd</sup> treatment T5	Negative	Negative	Negative	Negative	-	<1
Control	T5	Negative	Negative	Negative	Negative	-	-
Test cycle M-5							
BIO-SEA	2 <sup>nd</sup> treatment T5	Negative	Negative	Negative	Negative	-	<1
Control	T5	Negative	Negative	Negative	Negative	-	-

Table C-19 Primary production (DPM)

Facility	Position	FR1	FR2	FR3	Average	SD
Test cycle M-1						
Control/ BIO-SEA	Inlet	1,793.6	2,185.1	2,382.6	2,120.4	299.78
BIO-SEA	1 <sup>st</sup> treatment T0	15.7	16.7	15.7	16.0	0.56
BIO-SEA	2 <sup>nd</sup> treatment T5	0.22	3.85	0.51	1.52	2.02
Control	T5	191.3	217.5	194.4	201.0	14.31
Test cycle M-2						
Control/ BIO-SEA	Inlet	1,793.6	2,185.1	2,382.6	2,120.4	299.78
BIO-SEA	1 <sup>st</sup> treatment T0	23.3	17.1	15.6	18.7	4.11
BIO-SEA	2 <sup>nd</sup> treatment T5	7.58	1.59	3.05	4.07	3.12
Control	T5	191.3	217.5	194.4	201.0	14.31
Test cycle M-3						
Control/ BIO-SEA	Inlet	4,304.4	3,870.6	4,486.7	4,220.6	316.49
BIO-SEA	1 <sup>st</sup> treatment T0	19.2	11.3	14.9	15.1	3.96
BIO-SEA	2 <sup>nd</sup> treatment T5	0.51	2.38	0.47	1.12	1.09
Control	T5	1,499.4	1,463.6	1,410.1	1457.7	44.95
Test cycle M-4						
Control/ BIO-SEA	Inlet	1,202.7	1,056.0	1,116.5	1,125.1	73.72
BIO-SEA	1 <sup>st</sup> treatment T0	4.03	38.0	17.1	19.7	17.2
BIO-SEA	2 <sup>nd</sup> treatment T5	3.05	6.18	6.45	5.23	1.89
Control	T5	757.7	672.3	722.9	717.6	42.93
Test cycle M-5						
Control/ BIO-SEA	Inlet	1,202.7	1,056.0	1,116.5	1,125.1	73.72
BIO-SEA	1 <sup>st</sup> treatment T0	4.27	0.05	6.37	3.56	3.21
BIO-SEA	2 <sup>nd</sup> treatment T5	4.49	1.07	3.9	3.15	1.83
Control	T5	757.7	672.3	722.9	717.6	42.93

Table C-20 Number of organisms  $\geq 10$  and  $< 50 \mu\text{m}$  (Lugol's solution) (organisms/mL)

Facility	Position	FR1	FR2	FR3	Average	SD	IMO
Test cycle M-1							
Control/ BIO-SEA	Inlet	1,658	2,726	2,184	2,189	534	>1,000
Test cycle M-2							
Control/ BIO-SEA	Inlet	1,658	2,726	2,184	2,189	534	>1,000
Test cycle M-3							
Control/ BIO-SEA	Inlet	1,889	2,158	2,116	2,054	145	>1,000
Test cycle M-4							
Control/ BIO-SEA	Inlet	1,886	1,729	1,767	1,794	82	>1,000
Test cycle M-5							
Control/ BIO-SEA	Inlet	1,886	1,729	1,767	1,794	82	>1,000

Table C-21 Number of organisms  $\geq 10$  and  $< 50 \mu\text{m}$  (MPN) in brackish water (organisms/mL)

Facility	Position	FR1	FR2	FR3	Average	SD	IMO
Test cycle M-1							
Control/ BIO-SEA	Inlet	5,400	16,000	1,300	7,567	7,586	>1,000
BIO-SEA	2 <sup>nd</sup> treatment T5	<0.18	0.45	0.20	0.28	0.15	<10
Control	T5	920	540	350	603	290.23	>100
Test cycle M-2							
Control/ BIO-SEA	Inlet	5,400	16,000	1,300	7,567	7,586	>1,000
BIO-SEA	2 <sup>nd</sup> treatment T5	<0.18	<0.18	0.20	0.19	0.01	<10
Control	T5	920	540	350	603	290.23	>100
Test cycle M-3							
Control/ BIO-SEA	Inlet	16,000	5,400	16,000	12,467	6,120	>1,000
BIO-SEA	2 <sup>nd</sup> treatment T5	0.20	<0.18	<0.18	0.19	0.01	<10
Control	T5	>1,600	>1,600	1,600	>1,600	0.00	>100
Test cycle M-4							
Control/ BIO-SEA	Inlet	3,500	2,400	3,500	3,133	635	>1,000
BIO-SEA	2 <sup>nd</sup> treatment T5	<0.18	<0.18	<0.18	<0.18	0.00	<10
Control	T5	>1,600	>1,600	1,600	>1,600	0.00	>100
Test cycle M-5							
Control/ BIO-SEA	Inlet	3,500	2,400	3,500	3,133	635	>1,000
BIO-SEA	2 <sup>nd</sup> treatment T5	<0.18	<0.18	<0.18	0.18	0.00	<10
Control	T5	>1,600	>1,600	1,600	>1,600	0.00	>100

Table C-22 Number of organisms  $\geq 50 \mu\text{m}$  (organisms/ $\text{m}^3$ )

Facility	Position	FR1	FR2	FR3	Average	SD	IMO
Test cycle M-1							
Control	Inlet	137,760	137,147	119,173	131,360	10,558	>100,000
BIO-SEA	Inlet	134,200	147,920	114,813	132,311	16,634	>100,000
BIO-SEA	1 <sup>st</sup> treatment T0	26.7	10.0	16.7	17.8	8.39	-
BIO-SEA	2 <sup>nd</sup> treatment T5	0.00	0.00	0.00	0.00	0.00	<10
Control	T5	42,510	41,253	40,612	41,459	966	>100
Test cycle M-2							
Control	Inlet	137,760	137,147	119,173	131,360	10,558	>100,000
BIO-SEA	Inlet	101,640	112,327	117,413	110,460	8,051	>100,000
BIO-SEA	1 <sup>st</sup> treatment T0	6.67	23.3	3.33	11.1	10.7	-
BIO-SEA	2 <sup>nd</sup> treatment T5	0.00	0.00	0.00	0.00	0.00	<10
Control	T5	42,510	41,253	40,612	41,459	966	>100
Test cycle M-3							
Control	Inlet	101,627	130,240	97,240	109,702	17,921	>100,000
BIO-SEA	Inlet	99,013	97,745	119,947	105,569	12,468	>100,000
BIO-SEA	1 <sup>st</sup> treatment T0	20.0	20.0	6.67	15.6	7.70	-
BIO-SEA	2 <sup>nd</sup> treatment T5	0.00	0.00	0.00	0.00	0.00	<10
Control	T5	36,433	20,826	27,699	28,319	7,822	>100
Test cycle M-4							
Control	Inlet	198,000	183,300	126,000	169,100	38,042	>100,000
BIO-SEA	Inlet	142,667	211,600	206,500	186,922	38,411	>100,000
BIO-SEA	1 <sup>st</sup> treatment T0	10.0	10.0	25.0	15.0	8.66	-
BIO-SEA	2 <sup>nd</sup> treatment T5	0.00	0.00	0.00	0.00	0.00	<10
Control	T5	50,520	50,527	34,340	45,129	9,343	>100
Test cycle M-5							
Control	Inlet	198,000	183,300	126,000	169,100	38,042	>100,000
BIO-SEA	Inlet	210,000	219,600	225,400	218,333	7,778	>100,000
BIO-SEA	1 <sup>st</sup> treatment T0	7.50	7.50	2.50	5.83	2.89	-
BIO-SEA	2 <sup>nd</sup> treatment T5	0.00	0.00	0.00	0.00	0.00	<10
Control	T5	50,520	50,527	34,340	45,129	9,343	>100



## **A P P E N D I X   D**

Certificate of compliance, ISO 9001 certificate, accreditation and  
GLP authorisation

COPY

Certificate no: **DS/I093222**  
Page 1 of 1



## Certificate of Compliance

Office: **Lloyd's Register EMEA**  
**Copenhagen Design Support Centre, Statutory Section**  
**Strandvejen 104A, 2nd floor**  
**DK-2900 Hellerup**  
**Denmark**

Date: **24 June 2010**

This certificate is issued to **DHI Maritime Technology Evaluation Facility at Hundested, Denmark**

### **DHI Maritime Technology Evaluation Facility at Hundested, Denmark**

The Document(s) listed in paragraph 1 of the appendix have been examined for compliance with:

- Resolution MEPC.174(58), Annex part 2

and are found to comply from quality assurance and quality control aspects subject to the following:

- 1.1. It is required to maintain full and accurate log files in order to demonstrate correct quality measures
- 1.2. The Quality Assurance Project Plan is a project specific document and should as such be subject to review and commenting prior to each project start-up.
- 1.3. This design appraisal document is to be kept together with quality management plan.
- 1.4. Subject certificate is valid until 23 June 2015.

1. The documents listed below have been examined

Drawing No.	Rev.	Title	Status	Date
<b>Date: 23 June 2010</b>	<b>1</b>	<b>Quality Management Plan</b>	<b>B</b>	<b>24 June 2010</b>

2. The documents listed below have been considered together with the submitted documents in the appraisal

Drawing No.	Rev.	Title
<b>11806056</b>	<b>02</b>	<b>Quality Assurance Project Plan</b>

#### Appraisal Status Key

**B** Examined and found to comply with §2.1, Part 2 of the annex of IMO Resolution MEPC 174 (58)

*Martin Schabert*  
Martin Schabert  
Statutory Department  
Copenhagen Design Support Centre  
Surveyor to Lloyd's Register EMEA



A member of the Lloyd's Register Group

Lloyd's Register, its affiliates and subsidiaries and their respective officers, employees or agents are, individually and collectively, referred to in this clause as the 'Lloyd's Register Group'. The Lloyd's Register Group assumes no responsibility and shall not be liable to any person for any loss, damage or expense caused by reliance on the information or advice in this document or howsoever provided, unless that person has signed a contract with the relevant Lloyd's Register Group entity for the provision of this information or advice and in that case any responsibility or liability is exclusively on the terms and conditions set out in that contract.



---

# DET NORSKE VERITAS

---

## MANAGEMENT SYSTEM CERTIFICATE

---

Certificate No. 109333-2012-AQ-DEN-DANAK

*This is to certify that*

**DHI Group**

*has been found to conform to the management system standard:*

**DS/EN ISO 9001:2008**

*This certificate is valid for the following product or service ranges:*

**Consulting, software, research & development and laboratory testing, analysis & products  
within the area of water, environment & health**

Locations included in the certification will appear in the appendix.

*This certificate is valid until:*

**2015-01-10**

*The audit has been performed under the  
supervision of:*

**Jan Carsten Schmidt**  
*Lead Auditor*



*Place and date:*

**Hellerup, 2012-01-10**

**DET NORSKE VERITAS,  
BUSINESS ASSURANCE, DANMARK A/S**

**Jens Peter Høiseth**  
*Managing Director*

Lack of fulfilment of conditions as set out in the Certification Agreement may render this certificate invalid.



---

# DET NORSKE VERITAS

---

## APPENDIX TO CERTIFICATE

---

This appendix refers to certificate no. 109333-2012-AQ-DEN-DANAK

### DHI Group

Locations included in the certification are as follows:

Site Address	Scope:
Agern Allé 5 2970 Hørsholm, Denmark	Consulting, MIKE© by DHI Software Development, Sales & Support, Solutions Software Development, Research, Development & Innovation and Laboratory Analysis, Testing & Products
INCUBA Science Park, Gustav Wieds Vej 10 8000 Århus, Denmark	Consulting, Solutions Software Development and Research, Development & Innovation

*This certificate is valid until:*

2015-01-10

*The audit has been performed under the supervision of:*

Jan Carsten Schmidt  
*Lead Auditor*



*Place and date:*

Hellerup, 2012-01-10

DET NORSKE VERITAS,  
BUSINESS ASSURANCE, DANMARK A/S

Jens Peter Høiseth  
*Managing Director*

Lack of fulfilment of conditions as set out in the Certification Agreement may render this certificate invalid.

Company: **DHI**  
**Agern Allé 5**  
**DK-2970 Hørsholm**  
Registration number: **26**  
Valid: **04-07-2011 to 31-07-2015**

Scope:

**Testing**

**Product**

- **Biological items for testing**
- **Chemicals, chemical products, cosmetics, fertilizers, paints**
- **Environmental samples: Air, water, soil, waste**
- **Construction products**

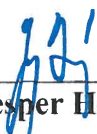
**Test Type**


- **Biological, biochemical testing**
- **Chemical testing, Analytical chemical testing**
- **Radiochemistry, radiation**
- **Sampling, laboratories accredited for sampling**

Testing is performed according to the current list of test methods approved by DANAK.

The company complies with the criteria in EN ISO/IEC 17025:2005 – General requirements for the competence of testing and calibration laboratories and demonstrates technical competence for the defined scope and the operation of a quality management system (refer joint ISO-ILAC-IAF Communiqué dated January 2009, [www.danak.dk](http://www.danak.dk)).

Issued July 4<sup>th</sup> 2011

  
**Jesper Høy**

  
**Kirsten Jebjerg Andersen**

COPY

DANAK

## GOOD LABORATORY PRACTICE

### STATEMENT OF COMPLIANCE

Laboratory inspection and study audits for compliance with the OECD Principles for Good Laboratory Practice were carried out at

Laboratory: DHI

on date: 25<sup>th</sup> March 2010 plus 7<sup>th</sup> and 9<sup>th</sup> April 2010

The laboratory inspection and study audits have been carried out in accordance with the regulation settled in Order No. 906 of 14<sup>th</sup> September 2009 from the Danish Ministry of Environment. The laboratory has been monitored for GLP Compliance within the following scope:

Type of products:


- *Industrial chemicals*
- *Pesticides*
- *Biocides*

Type of tests:

- *Environmental toxicity studies on aquatic and terrestrial organisms.*
- *Studies of behaviour in water, soil and air, bioaccumulation*

The laboratory was found to be operating in compliance with the OECD Principles of Good Laboratory Practice.

Date: 2<sup>nd</sup> December 2010

  
Jesper Høy  
Managing director, DANAK

  
Kirsten Jebjerg Andersen  
GLP inspector, DANAK